**Safety Assessment of Natural Products in Malaysia: Current Practices, Challenges, and New Strategies**

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**Abstract**

The belief that natural products are inherently safe is a primary reason for consumers to choose traditional medicines and herbal supplements for health maintenance and disease prevention. Unfortunately, some natural products on the market have been found to contain toxic compounds, such as heavy metals and microbes, as well as banned ingredients such as aristolochic acids. It shows that the existing regulatory system is inadequate and highlights the importance of thorough safety evaluations. In Malaysia, the National Pharmaceutical Regulatory Agency is responsible for the regulatory control of medicinal products and cosmetics, including natural products. For registration purpose, the safety of natural products is primarily determined through the review of documents, including monographs, research articles and scientific reports. One of the main factors hampering safety evaluations of natural products is the lack of toxicological data from animal studies. However, international regulatory agencies such as the European Food Safety Authority and the United States Food and Drug Administration are beginning to accept data obtained using alternative strategies such as non-animal predictive toxicological tools. Our paper discusses the use of state-of-the-art techniques, including chemometrics, in silico modelling and omics technologies and their applications to the safety assessments of natural products.

Keywords: chemometrics; herbal medicines; in silico; predictive toxicology; omics; safety assessments

**Abbreviations**

(Q)SAR, (quantitative) structure-activity relationship

3R, replacement, reduction, and refinement

AAs, aristolochic acids

AB, alkenylbenzene

ADR, adverse drug reactions

CAM, complementary and alternative medicines

DCA, Drug Control Authority

DRGD, Drug Registration Guidance Document

EFSA, European Food Safety Authority

FDA, Food and Drug Administration

GMP, Good manufacturing practice

HPLC, high performance liquid chromatography

IARC, International Agency for Research on Cancer

JECFA, Expert Committee on Food Additives

NPRA, National Pharmaceutical Regulatory Agency

PA, pyrrolizidine alkaloids

PBK, physiologically-based kinetic

WHO, World Health Organization

**Introduction**

The use of herbal medicines in Malaysia dates back to ancient times and continues to the current day. Herbal medicines, which include herbs, herbal materials, herbal preparations, and finished herbal products containing plants or parts of plants as the active ingredients, are the core component of traditional medicines in many Malaysian cultures including Malay folk medicine, traditional Chinese medicine, Ayurvedic medicine and homeopathy. According to a recent report by the World Health Organization (WHO), about 9 out of 30 million Malaysians have used traditional medicines to prevent or treat illnesses (1). Due to the popularity of natural products, such as plant food supplements and herbal medicines, the Malaysian herbal industry has grown substantially in recent years, with the market value is expected to reach MYR 32 billion by 2020, representing an annual growth rate of 8-15% (2).

The herbs used in herbal medicines are rich in phytochemicals such as carotenoids, phenolics, alkaloids, nitrogenous compounds, and organosulfur compounds (3). While many phytochemicals have therapeutic properties, such as anticancer, antitumor, and antioxidant activity, some of these compounds also exhibit toxic effects, such as genotoxic, carcinogenic, hepatotoxic, and nephrotoxic effects (4). The notion that herbal medicines are always safe and harmless because they are natural is therefore incorrect, as previous studies have demonstrated that herbal medicines can induce mild to severe acute or spontaneous adverse drug reactions (ADR). For example, valerian (*Valeriana officinalis*) causes extreme fatigue, black cohosh (*Actaea racemosa*) causes upper abdominal pain and hepatoxicity, and concomitant use of *Ginkgo biloba* with the anticoagulant drug warfarin can result in haemorrhage (5). A recent update by the Malaysian National Pharmaceutical Regulatory Agency (NPRA), reported that products containing lingzhi or reishi (*Ganoderma lucidum*), a medicinal mushroom, caused 9 kidney-related and 2 liver-related ADR, with a total of 8 patients requiring hospitalisation (6). In addition, there are numerous reports of the contamination of herbal medicine with toxic compounds. These contaminants may have occurred naturally or from microorganisms or environmental contamination (7, 8).

The use of herbal medicines has also been associated with chronic toxicity, including carcinogenicity, teratogenicity, hepatotoxicity, nephrotoxicity, and others (7, 9). Some naturally occurring phytochemicals, such as pyrrolizidine alkaloids (PAs) and alkenylbenzene (AB) are genotoxic and carcinogenic (10). Exposure to PAs has been associated with acute toxic effects on the liver, resulting in hepatomegaly, ascites, and veno-occlusive disease , as well as chronic liver toxicities (11). International Agency for Research on Cancer (IARC) has classified lasiocarpine, monocrotaline, and riddelliine (congeners of PAs) and safrole (a congener of AB) to be ‘possibly carcinogenic to humans’ (2B) (12). In relation to the matter, the German Federal Institute for Risk Assessment (BfR) recommended that the content of PAs in food to be kept at lowest possible level (13).

In general, the regulatory requirements for safety assessments of these products are less stringent compared to synthetic pharmaceuticals. Most herbal medicines and plant food supplements have never been thoroughly evaluated for safety before being allowed into market (14). The 2019 WHO Global Report on Traditional and Complementary Medicine (1) reports that 38 Member States have no regulatory requirements for safety assessments of herbal medicines, 35 Member States consider that the safety of a herbal medicine is based on the absence of adverse effects when used traditionally, and 26 Member States accept safety data from scientific research on similar products. In many countries, herbal medicines with long history of widespread use are considered safe when correctly administered in the usual doses (15). In Europe, the safety status of herbal medicines is based on the absence of known adverse effects from long-standing traditional use. In the United States of America, herbal and dietary supplements are exempt from rigorous review by the Food and Drug Administration (FDA), because they are considered as a low-risk products (16, 17). In Malaysia, toxicological studies are not required for ingredients that have been used without safety concerns unless the product contains a new active ingredient or combination of ingredients (18). Table 1 provides an overview of the safety assessments of herbal medicines in Australia, Europe and United Kingdom, India, Japan, and the United States of America.

Our paper describes the pathways for registration and safety assessment of natural products in Malaysia, discusses the challenges of the current approach and presents new methodologies in strategies for safety assessments that have been discussed in the recent literatures.

**Regulation of Natural Products and Health Supplements in Malaysia**

*Registration and Safety Assessment*

Under the Malaysian Control of Drug and Cosmetics Regulation 1984, natural products which include traditional medicines, finished herbal products, herbal remedies, homeopathic medicines, and natural products with therapeutic claims must be registered with the Drug Control Authority (DCA). The NPRA, which acts as the secretariat for the DCA, has published Drug Registration Guidance Document (DRGD), an important reference for the registration of medicinal products, covering quality control, inspection, licencing, and post-registration activities (18). Vitamins and minerals, in the form of extracts or isolates, derived from natural sources including plants, animals or minerals are classified as health supplements (18).

In general, each application received is subject to a pre-marketing assessment before an authorisation decision can be taken. Each application must be supported with evidence of safety and efficacy, based on the claims of the product. Official scientific documents such as standard references from textbooks, pharmacopoeias, monographs, or reference organisations are accepted. However, toxicological data (e.g., chronic toxicity studies) and clinical data are mandatory only for natural products and health supplements with disease reduction claim, but not for others. The DRGD includes guidelines, such as maximum levels of vitamins and minerals permitted in health supplements for adults, and botanical ingredients that are banned or prohibited from product formulations. For instance, all plants from the genus of Aristolochia is banned due to their potential nephrotoxicity and some plants from the genera Symphytum and Senecio are banned due to their potential hepatoxicity.

In Malaysia, natural products are subject to laboratory testing for microbial and heavy metal contamination prior approval as part of the safety assessments, and all products must comply to the limits specified in the DRGD. However, a recent change allows these tests to be performed by local panel laboratories or local manufacturer laboratories, whereas in the past they were performed by the NPRA’s laboratory (19). In addition, products containing botanicals listed in List A (botanicals known or suspected to contain aristolochic acids (AAs), such as *Asarum canadense* Linn. and *Bragantia wallichii* R. Br) and List B (botanicals which may be adulterated with AAs, such as *Asarum sieboldii* Miq and *Asarum heterotropoides* F. Schmidt) must be tested for presence of AAs. The overview of the regulatory pathways for natural products in Malaysia is shown in Fig 1.

Manufacturers of natural products and health supplements must demonstrate compliance with Good Manufacturing Practice (GMP) as outlined in the Guideline on Good Manufacturing Practice for Traditional Medicines and Herbal Supplements (20). One of the GMP requirements for herbal medicines is the correct identification of the source of the plant species and the plant part(s) (21). For registration purpose, manufacturers must ensure that their raw materials used are identified or authenticated and the details must be included in the certificate of analysis of the active ingredients (18, 19). The NPRA also actively monitors registered products in the market as part of its continuous post-marketing surveillance activities, and failure to meet quality and safety standards will result in cancellation of product registration and removal of the product from the market. In addition, the NPRA handles all complaints related to quality, efficacy, labelling and packaging of products, and deals with the ADR reports through the National ADR Monitoring Centre. The system enables effective regulatory actions to be taken against potentially harmful products, thereby minimising the risks to consumers.

*Challenges and Issues with the Current System*

While provisions or regulations regarding natural products and health supplements have already been established in Malaysia, various studies show that herbal products with various types of contaminants are still entering the market (Table 2). Heavy metals are toxic even at low concentrations, e.g., mercury, arsenic, cadmium and lead can cause mutagenic effects even at low levels (22). On the other hand, contamination of herbal medicines with pathogens including Salmonella, Eschericia coli and Pseudomonas aeruginosa has caused serious health issues worldwide, especially among immunocompromised consumers (23). Certain strains of *Aspergillus* fungi such as *A. flavus* and *A. nomius* produce toxic secondary metabolites , which are carcinogenic to humans (24).In addition, significant levels of AAs were found in the analysis of 18 plants food supplements obtained through global online purchase (AAI: 2.1 – 594.8 μg/g, AAII: 0.6 – 235.3 μg/g) (25). These findings clearly show that despite the ban, AAs containing products are still available on the market.

Fig. 2 shows that there has been a tremendous increase in the number of ADR reports of traditional medicines, vitamins, herbal supplements, and minerals in Malaysia. However, reports of under-reporting of ADR of herbal products by health professionals and consumers suggest that the actual number of cases may be even greater (27). A retrospective study by Rahman and Aziz reported 242 (26%) serious adverse drug reactions (ADRs), including 36 deaths as a result of complementary and alternative medicines use (27). From 2010 to 2016, 140 cases of acute kidney injury were associated with the consumption of traditional and complementary medicines (28). Although both studies show that the majority of adverse reactions are caused by unregistered natural products, there are still reports of adverse reactions caused by licensed products that need to be addressed.

While most herbal medicines are evaluated for safety and efficacy based on information found in monographs, research articles, and scientific reports, the method is inadequate because there are few scientific studies and clinical trials of high methodological quality (15). Most of the pharmacological and toxicological studies could not be verified due to the difference in extraction methods as different solvents may affect the composition of the test material and thus affect the results (29). For the same reason, the actual toxicity of herbal medicine is not known, and the assumption of safety of herbal medicines based on long-standing traditional use is not justified. Moreover, the recent reform whereby pre-registration testing of natural products is now carried out by the manufacturer rather than the regulatory authority (NPRA) may again raise questions about the validity and credibility of the test results. Consequently, there is an urgent need for more comprehensive safety assessments of herbal products, that include in silico modelling, chemical fingerprinting, chemometric methods, and omics-based approaches that minimise the risk associated with the use of herbal products.

**New Strategies for Safety Evaluation of Natural Products**

*Chemical fingerprinting and chemometric evaluations*

Some herbal products are formulated to have "synergistic" effects by combining many different plant species. It is especially common in traditional Chinese medicine, where most herbal medicines prescribed by the practitioners are polyherbal and may even contain animal parts and mineral substances to form *fu-fang* or multi-item concoction (30). Therefore, the chemical composition is extremely diverse, with hundreds of known and unknown compounds present within the same product. Currently, the quality and authenticity of a plant species is assessed by chromatographic determination of one or two pharmacologically active constituents or biomarkers. However, it is often insufficient to provide a complete fingerprint, especially when the composition is too complex, as in the case of polyherbs (31). Failure to authenticate a plant species in a product can lead to serious hazardous effects to the consumers, such as adulteration with related species or banned substances.

Herbal profiling is no easy task. The integration of high-tech chromatography instruments (e.g., ultra-high performance liquid chromatography (HPLC), reversed phase ion pairing HPLC, size exclusion chromatography) and mass spectrometers (e.g., UV-diode array detector, electrospray ionisation-mass spectrometry), or hyphenated technique, however is superior to conventional methods because it provides rapid and highly effective separation, higher selectivity and precision, elimination of instrumental interferences, and correction of the retention time. The resulting data which consist of a chromatographic fingerprint and spectral data, can be analysed using chemometrics, a discipline of chemistry that employs mathematical and statistical methods. Using chemometrics, the complicated fingerprint data generated can be classified according to the pattern, and then calibrated to remove unwanted data from instrument errors, allowing useful information to be extracted (31).

*Predictive toxicology using computational method.*

The cumbersome, time-consuming and expensive procedures are the reasons for the lack of in vivo or animal toxicity studies for herbal medicines. Moreover, the exorbitant use of animals for toxicity studies is strongly discouraged under the Replacement, Reduction, and Refinement (3Rs) initiative. To overcome the issues, researchers have explored the use of computational methods or in silico methods to predict the in vivo toxicity of chemical compounds (32, 33). Generally, in silico methods use mathematical equations to create simulation models that can predict the activity of a compound in biological system, based on existing knowledge about the compound (34). To develop the model, data such as the physicochemical properties and biological activities of the chemicals will serve as input to computer software that performs regression analysis to develop the mathematical equations or model that represents the effect in vivo. The developed model is then validated against published data and can then be used to accurately predict various toxicological endpoints. In silico methods, such as (quantitative) structure-activity relationship [(Q)SAR], physiologically-based kinetic (PBK) modelling, and molecular docking are used in drug discovery, pesticide toxicity estimation, and safety assessment of food ingredients and cosmetics (32, 33). In addition, several international regulatory agencies have also begun to use in silico methods for risk assessments (35, 36) that require exposure assessment data (37).

(Q)SAR approach is built on the basis that the biological activity of a compound is related to its chemical structure. Prediction results from (Q)SAR software (e.g., MDL-QSAR, DEREK, and Leadscope) are checked for validity against published experimental results. On the other hand, PBK modelling incorporates in vitro data and in silico kinetic data to simulate bioactivity and toxicity in vivo. PBK model-reverse dosimetry enables quantitative in vitro-in vivo extrapolation (QIVIVE) which translates concentration-response curves from in vitro studies into in vivo dose-response curves by incorporating species toxicokinetic and toxicodynamic data. Similar to (Q)SAR, there are a variety of platforms for PBK modelling, such as Berkeley Madonna, acslXtreme, Simcyp, and MATLAB (38). Another in silico method that has been used in predictive toxicology is molecular modelling and docking. This technique uses the knowledge of molecular structure and properties to model the behaviour of a molecule and predict binding affinity to the active site of a receptor. In molecular modelling, molecular structures are represented numerically, and its behaviour is simulated by equations. For risk assessment process, simulations based on molecular modelling and docking can provide supportive data when information on interaction with targets is lacking (34, 39).

*Toxicogenomic in predictive toxicology*

Toxicogenomic is the integration of toxicological and molecular science studies that examine changes in mRNA expression, protein expression, and metabolite profiles following exposure to a contaminant (40). The use of omics methods in the safety assessments of herbal medicines and traditional Chinese medicines have been reviewed (41-44). In general, a typical metabolomics analysis begins with the collection on samples, such as cell cultures or biological samples such as urine, serum, plasma, and tissue. The samples are subjected to extraction and fractionation to separate the proteins, peptides, and metabolites, using chromatography or electrophoresis techniques. Subsequently, the proteins and metabolites in the samples are quantified using nuclear magnetic resonance or mass spectrometry, the latter being more common for proteomics and metabolomics analysis. On the other hand, other methods such as DNA microarrays, oligonucleotide-based array and complementary DNA arrays are also used for genomics and transcriptomics analysis, providing information on mRNA or gene expression, or DNA sequence (44). The data generated are typically large and complex, requiring pre-processing using multivariate statistical methods to improve the representation of the data, to allow interpretation and elucidation of the mode of action of toxicity or biomarker identification (35, 42).

Table 3 provides an overview of the advantages and limitations of each the new strategies. The alternative methods can accurately analyse complex mixtures of chemicals, which is extremely important for natural products especially in Malaysia as they are usually polyherbal. Moreover, these methods eliminate the problems associated with in vitro and in vivo experiments, for instance, long duration of experiments, significant financial and human resources, and ethical issues in animal testing.

**Conclusion**

The belief that natural products are inherently safe and effective is a primary reason for consumers to choose traditional medicines and herbal supplements for health maintenance and disease prevention. Unfortunately, these natural products are not adequately regulated and may contain toxic ingredients. In Malaysia, products with banned ingredients such as AAs, contaminated with microbes and heavy metals. still find their way into the market. Long-term exposure to toxic ingredients is not only harmful to vulnerable populations such as the elderly and immunocompromised, but also significantly increases the risk of developmental disorders in infants and children, reproductive complications, and chronic diseases such as cancer in other populations. To ensure the safety and quality of natural products, the Malaysian regulatory authority must ensure that manufacturers comply with regulations, i.e., implement and adhere to Good Agricultural and Collection Practices and GMP. Next, safety assessments for natural products need to be supported by newer strategies with advanced analytical instruments and computational techniques that include authentication of plant materials with chromatographic fingerprinting and chemometric tools, and predictive computational toxicology. This will require extensive training and ongoing education of the laboratory analysts and regulators in the field. Collaboration with international agencies that have expertise in these areas can also be a starting point. Despite the strengths and weaknesses of each of the new strategies, these techniques offer much greater benefits than the conventional experimental methods. In addition, integration of the methods would provide invaluable and comprehensive data on the properties of the chemical substances and their effects on the nature of toxicity and development of toxicity endpoints or diseases, thus creating a rigorous methodology for assessing the safety of natural products.

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**References**

1. WHO. WHO global report on traditional and complementary medicine 2019. Geneva, Switzerland2019. 228- p.

2. BERNAMA. Malaysian herbal industry poised to hit RM32 bln market value in 2020. BERNAMA. 2019.

3. Liu R H. Potential Synergy of Phytochemicals in Cancer Prevention: Mechanism of Action. The Journal of Nutrition. 2004;134(12):3479S-85S.

4. Chopra A, Doiphode VV. Ayurvedic medicine. Core concept, therapeutic principles, and current relevance. Med Clin North Am. 2002;86(1):75-89, vii.

5. Hoban CL, Byard RW, Musgrave IF. Analysis of spontaneous adverse drug reactions to echinacea, valerian, black cohosh and ginkgo in Australia from 2000 to 2015. Journal of Integrative Medicine. 2019;17(5):338-43.

6. Reduzan NAM, Chet LS. NPRA Updates on Reports of Kidney-Related and Liver-Related Adverse Reactions after Consumption of Ganoderma (Lingzhi / Reishi) Products. NPRA; 2020.

7. Ekor M. The growing use of herbal medicines: Issues relating to adverse reactions and challenges in monitoring safety. Frontiers in Neurology. 2014;4 JAN(January):1-10.

8. Bateman J, Chapman RD, Simpson D. Possible toxicity of herbal remedies. Scott Med J. 1998;43(1):7-15.

9. Guldiken B, Ozkan G, Catalkaya G, Ceylan FD, Ekin Yalcinkaya I, Capanoglu E. Phytochemicals of herbs and spices: Health versus toxicological effects. Food and Chemical Toxicology. 2018;119(May):37-49.

10. van d Berg S J P L, Restani P, Boersma M G, Delmulle L, Rietjens I M C M. Levels of Genotoxic and Carcinogenic Ingredients in Plant Food Supplements and Associated Risk Assessment. Food and Nutrition Sciences. 2011;02(09):989-1010.

11. Wiedenfeld H. Plants containing pyrrolizidine alkaloids: Toxicity and problems. Food Additives and Contaminants - Part A Chemistry, Analysis, Control, Exposure and Risk Assessment. 2011;28(3):282-92.

12. IARC. Agents Classified by the IARC Monographs, Volumes 1–127 2018 [updated 26th June 2020. Available from: <https://monographs.iarc.fr/agents-classified-by-the-iarc/>.

13. Ma C, Liu Y, Zhu L, Ji H, Song X, Guo H, et al. Determination and regulation of hepatotoxic pyrrolizidine alkaloids in food: A critical review of recent research. Food and Chemical Toxicology. 2018;119:50-60.

14. Schilter B, Andersson C, Anton R, Constable A, Kleiner J, O'Brien J, et al. Guidance for the safety assessment of botanicals and botanical preparations for use in food and food supplements. Food Chem Toxicol. 2003;41(12):1625-49.

15. Sahoo N, Manchikanti P, Dey S. Herbal drugs: Standards and regulation. Fitoterapia. 2010;81(6):462-71.

16. Alostad A H, Steinke D T, Schafheutle E I. International Comparison of Five Herbal Medicine Registration Systems to Inform Regulation Development: United Kingdom, Germany, United States of America, United Arab Emirates and Kingdom of Bahrain. Pharmaceutical Medicine. 2018;32(1):39-49.

17. Moreira R, Pereira DM, Valentão P, Andrade PB. Pyrrolizidine alkaloids: Chemistry, pharmacology, toxicology and food safety. International Journal of Molecular Sciences. 2018;19(6).

18. NPRA. Drug Registration Guidance Document 2nd ed 2016.

19. NPRA. Part 2: Regulatory Updates 2019-2020 and Strategic Plan 2021-2024 2020 [Available from: <https://www.npra.gov.my/images/PART_2_REGULATORY_UPDATES_130220_v1.pdf>.

20. NPRA. Guideline on Good Manufacturing Practice for Traditional Medicines and Herbal Supplements First ed 2008.

21. Blumenthal M. Herbal Monographs. The Journal of the American Botanical Council. 1997(40):30.

22. Shaban N S, Abdou K A, Hassan N E-H Y. Impact of toxic heavy metals and pesticide residues in herbal products. Beni-Suef University Journal of Basic and Applied Sciences. 2016;5(1):102-6.

23. Famewo E B, Clarke A M, Afolayan A J. Identification of bacterial contaminants in polyherbal medicines used for the treatment of tuberculosis in Amatole District of the Eastern Cape Province, South Africa, using rapid 16S rRNA technique. J Health Popul Nutr. 2016;35(1):27.

24. Tajkarimi M, Shojaee M, Yazdanpanah H, Ibrahim S. Aflatoxin in Agricultural Commodities and Herbal Medicine. 2011.

25. Abdullah R, Diaz L N, Wesseling S, Rietjens I M C M. Risk assessment of plant food supplements and other herbal products containing aristolochic acids using the margin of exposure (MOE) approach. Food Additives and Contaminants - Part A Chemistry, Analysis, Control, Exposure and Risk Assessment. 2017;34(2):135-44.

26. IARC. IARC Monograph 100A - Plants Containing Aristolochic Acid 2002.

27. S A Rahman S, Aziz Z. Complementary and alternative medicine: Pharmacovigilance in Malaysia and predictors of serious adverse reactions. Journal of Clinical Pharmacy and Therapeutics. 2020;45.

28. Lee F, Wong H, Tay J, Choong C, Mohamed Ali N, editors. Acute kidney injury related to traditional/complementary medicines of supplements in Malaysia. 1ST ASIA PACIFIC AKI CRRT 2017; 2017; Kuala Lumpur: Kidney International Reports

29. Wiesner J, Knöss W. Future visions for traditional and herbal medicinal Products - A global practice for evaluation and regulation? Journal of Ethnopharmacology. 2014;158(PART B):516-8.

30. Che C T, Wang Z J, Chow M S, Lam C W. Herb-herb combination for therapeutic enhancement and advancement: theory, practice and future perspectives. Molecules. 2013;18(5):5125-41.

31. Bansal A, Chhabra V, Rawal RK, Sharma S. Chemometrics: A new scenario in herbal drug standardization. J Pharm Anal. 2014;4(4):223-33.

32. Sripriya N, Ranjith Kumar M, Ashwin Karthick N, Bhuvaneswari, Udaya Prakash N. In silico evaluation of multispecies toxicity of natural compounds. Drug and Chemical Toxicology. 2019:1-7.

33. Arvidson K B, Valerio L G, Diaz M, Chanderbhan R F. In Silico Toxicological Screening of Natural Products. Toxicology Mechanisms and Methods. 2008;18(2-3):229-42.

34. Eweas A F. Advances in molecular modeling and docking as a tool for modern drug discovery. Der Pharma Chemica 2014;6(6):211-28.

35. Jordan S A, Cunningham D G, Marles R J. Assessment of herbal medicinal products: challenges, and opportunities to increase the knowledge base for safety assessment. Toxicol Appl Pharmacol. 2010;243(2):198-216.

36. Paini A, Leonard J A, Joossens E, Bessems J G M, Desalegn A, Dorne J L, et al. Next generation physiologically based kinetic (NG-PBK) models in support of regulatory decision making. Computational Toxicology. 2019;9:61-72.

37. Schilter B, Benigni R, Boobis A, Chiodini A, Cockburn A, Cronin MT, et al. Establishing the level of safety concern for chemicals in food without the need for toxicity testing. Regul Toxicol Pharmacol. 2014;68(2):275-96.

38. Rietjens I M, Louisse J, Punt A. Tutorial on physiologically based kinetic modeling in molecular nutrition and food research. Mol Nutr Food Res. 2011;55(6):941-56.

39. James R. Rabinowitz M-RG,Stephen B. Little, and Melissa A. Pasquinelli. Computational Molecular Modeling for Evaluating the Toxicity of Environmental Chemicals: Prioritizing Bioassay Requirements. Environmental Health Perspectives. 2008;16.

40. Oberemm A, Onyon L, Gundert-Remy U. How can toxicogenomics inform risk assessment? Toxicology and Applied Pharmacology. 2005;207(2, Supplement):592-8.

41. Ouedraogo M, Baudoux T, Stévigny C, Nortier J, Colet J-M, Efferth T, et al. Review of current and “omics” methods for assessing the toxicity (genotoxicity, teratogenicity and nephrotoxicity) of herbal medicines and mushrooms. Journal of Ethnopharmacology. 2012;140(3):492-512.

42. Duan L, Guo L, Wang L, Yin Q, Zhang C-M, Zheng Y-G, et al. Application of metabolomics in toxicity evaluation of traditional Chinese medicines. Chinese Medicine. 2018;13(1).

43. Pelkonen O, Duez P, Vuorela PM, Vuorela H. Toxicology of herbal products2017. 1-479 p.

44. Woo C S J, Lau J S H, El-Nezami H. Chapter 10 - Herbal Medicine: Toxicity and Recent Trends in Assessing Their Potential Toxic Effects. In: Shyur L-F, Lau ASY, editors. Advances in Botanical Research. 62: Academic Press; 2012. p. 365-84.

45. TGA. Australian regulatory guidelines for complementary medicines (ARGCM) ver 8.0. In: Health Do, editor. 2018.

46. Commission E. Directive 2004/24/EC of the European Parliament and of the Council of 31 March 2004 amending, as regards traditional herbal medicinal products, Directive 2001/83/EC on the Community code relating to medicinal products for human use. In: Agency EM, editor. Brussels2004.

47. Sahoo N, Manchikanti P. Herbal drug regulation and commercialization: an Indian industry perspective. J Altern Complement Med. 2013;19(12):957-63.

48. Organization CDSC. The Drugs and Cosmetics Act. 1940.

49. Maegawa H, Nakamura T, Saito K. Regulation of traditional herbal medicinal products in Japan. J Ethnopharmacol. 2014;158 Pt B:511-5.

50. Thakkar S, Anklam E, Xu A, Ulberth F, Li J, Li B, et al. Regulatory landscape of dietary supplements and herbal medicines from a global perspective. Regulatory Toxicology and Pharmacology. 2020;114:104647-.

51. 2016. Botanical Drug Development - Guidance for Industry. In: Services USDoHaH, editor. FDA.

52. Ang H H, Lee E L, Cheang H S. Determination of mercury by cold vapor atomic absorption spectrophotometer in Tongkat Ali preparations obtained in Malaysia. Int J Toxicol. 2004;23(1):65-71.

53. Ang H H, Lee K L. Contamination of mercury in tongkat Ali hitam herbal preparations. Food Chem Toxicol. 2006;44(8):1245-50.

54. Ang H H, Lee E L, Matsumoto K. Analysis of lead content in herbal preparations in Malaysia. Hum Exp Toxicol. 2003;22(8):445-51.

55. Ang H H, Lee K L, Kiyoshi M. Determination of lead in Smilax luzonensis herbal preparations in Malaysia. Int J Toxicol. 2005;24(3):165-71.

56. Ang H H, Lee KL, Kiyoshi M. Determination of lead in Smilax myosotiflora herbal preparations obtained in Malaysia. Int J Environ Health Res. 2004;14(4):261-72.

57. Ang H H. Lead contamination in Eugenia dyeriana herbal preparations from different commercial sources in Malaysia. Food Chem Toxicol. 2008;46(6):1969-75.

58. Ali N, Hashim N H, Saad B, Safan K, Nakajima M, Yoshizawa T. Evaluation of a method to determine the natural occurrence of aflatoxins in commercial traditional herbal medicines from Malaysia and Indonesia. Food Chem Toxicol. 2005;43(12):1763-72.

59. Bustami N A, Ho Y B, Tan C K, Aris A Z, Tan E S S. Consumption of Manjakani Among Postpartum Mothers and Risk of Heavy Metal Contamination Malaysian Journal of Medicine and Health Sciences. 2020;16(2):75-82.

60. Abu Bakar N F, Mohamad Zin N, Long Tuan Kechik T S M, Santhanam J. Commercial Herbal Slimming Products: Evaluation of Heavy Metals and Microorganism Contamination at Different Batch Production. Jurnal Sains Kesihatan Malaysia. 2017;15(01):7-18.

61. Gad H A, El-Ahmady S H, Abou-Shoer M I, Al-Azizi M M. Application of chemometrics in authentication of herbal medicines: a review. Phytochem Anal. 2013;24(1):1-24.

62. Jing D, De-guang W, Lin-fang H, Shi-lin C, Min-jian Q. Application of chemometrics in quality evaluation of medicinal plants. Journal of Medicinal Plants Research. 2011;5:4001-8.

63. Guo L, Mei N, Dial S, Fuscoe J, Chen T. Comparison of gene expression profiles altered by comfrey and riddelliine in rat liver. 2007;8(Suppl 7):S22.

64. Mei N, Guo L, Liu R, Fuscoe JC, Chen T. Gene expression changes induced by the tumorigenic pyrrolizidine alkaloid riddelliine in liver of Big Blue rats. BMC Bioinformatics. 2007;8(S7):S4.

65. Argueso C T, Assmann S M, Birnbaum K D, Chen S, Dinneny J R, Doherty C J, et al. Directions for research and training in plant omics: Big Questions and Big Data. Plant Direct. 2019;3(4):e00133.

66. Neves B J, Braga R C, Melo-Filho C C, Moreira-Filho J T, Muratov E N, Andrade C H. QSAR-Based Virtual Screening: Advances and Applications in Drug Discovery. Frontiers in Pharmacology. 2018;9(1275).

67. Kruhlak N L. US FDA Experience in the Regulatory Application of (Q)SAR 2020 [Available from: <https://www.toxicology.org/groups/ss/CTSS/docs/SOT-CE-Presentation-Kruhlak_final_Updated2.pdf>.

68. Sayada Reemsha Kazmi RJ, Myeong-Sang Yu, Chanjin Jung, Na D. In silico approaches and tools for the prediction of drug metabolism and fate : A review. Computers in Biology and Medicine. 2019;106:54-64.

69. OECD. Guidance document on the characterisation, validation and reporting of Physiologically Based Kinetic (PBK) models for regulatory purposes. Series on Testing and Assessment No 331: OECD; 2021.

70. Hartmanshenn C, Scherholz M, Androulakis IP. Physiologically-based pharmacokinetic models: approaches for enabling personalized medicine. J Pharmacokinet Pharmacodyn. 2016;43(5):481-504.

71. Tan Y M, Worley R R, Leonard J A, Fisher J W. Challenges Associated With Applying Physiologically Based Pharmacokinetic Modeling for Public Health Decision-Making. Toxicol Sci. 2018;162(2):341-8.

72. Chen L, Ning J, Louisse J, Wesseling S, Rietjens IMCM. Use of physiologically based kinetic modelling-facilitated reverse dosimetry to convert in vitro cytotoxicity data to predicted in vivo liver toxicity of lasiocarpine and riddelliine in rat. Food and Chemical Toxicology. 2018;116(March):216-26.

73. Louisse J, Bosgra S, Blaauboer BJ, Rietjens IM, Verwei M. Prediction of in vivo developmental toxicity of all-trans-retinoic acid based on in vitro toxicity data and in silico physiologically based kinetic modeling. Arch Toxicol. 2015;89(7):1135-48.

74. Ning J, Chen L, Rietjens IMCM. Role of toxicokinetics and alternative testing strategies in pyrrolizidine alkaloid toxicity and risk assessment; state-of-the-art and future perspectives. Food and Chemical Toxicology. 2019;131:110572.

75. Mansouri K, Kleinstreuer N, Abdelaziz AM, Alberga D, Alves VM, Andersson PL, et al. CoMPARA: Collaborative Modeling Project for Androgen Receptor Activity. Environ Health Perspect. 2020;128(2):27002.

76. Mansouri K, Abdelaziz A, Rybacka A, Roncaglioni A, Tropsha A, Varnek A, et al. CERAPP: Collaborative Estrogen Receptor Activity Prediction Project. Environ Health Perspect. 2016;124(7):1023-33.

77. Huang SY, Zou X. Advances and challenges in protein-ligand docking. Int J Mol Sci. 2010;11(8):3016-34.

78. MADRAC. National Centre for Adverse Drug Reactions Monitoring Annual Report 2018 [Available from: <https://npra.gov.my/index.php/en/informationen/annual-reports/national-centre-for-adverse-drug-reaction-monitoring-annual-report>.

**Table 1: Overview of safety assessment of herbal medicines by different countries/regulatory authorities**

|  |  |  |
| --- | --- | --- |
| **Country (Regulatory Authority)** | **Description** | **References** |
| Australia (Therapeutic Goods Administration, TGA) | To demonstrate the safety of use of complementary medicines, the history of use, and scientific data (e.g., animal studies, expert reports, reviews, evaluation reports from other agencies) must be provided. Conventional studies involving animal and in vitro studies are not required if sufficient clinical and/or historical human evidence is available. Epidemiological studies on food or health supplements are also acceptable. However, lack or incomplete of evidence in humans and clinical data shall be supported by other studies such as repeat-dose toxicity, genotoxicity, carcinogenicity, reproductive and developmental toxicity, etc.  | (45) |
| United Kingdom and Europe Union (European Medicines Agency, EMA)  | The simplified registration pathway applies to traditional herbal medicinal products with a long tradition of medicinal use (i.e., established traditional use for at least 30 years, with at least of 15 years of medicinal use in the EU) whereby no clinical trials are required to demonstrate safety. However, specific eligibility criteria as outlined in the EU Directive 2004/24/EC (e.g., only medicines for oral, topical and inhalation administrations are authorised) must be met to enable this registration pathway.  | (46) |

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| **Table 1: Continued** |
| **Country (Regulatory Authority)** | **Description** | **References** |
| India (Ministry of AYUSH) | Clinical evidence is not required to support safety Ayurveda, Siddha and Unani (ASU) drugs manufactured as defined in Section 3(a) of the Drugs and Cosmetics Act, 1940 and drugs containing aqueous extract of the medicinal plants. On the other hand, safety and efficacy studies are mandatory for proprietary ASU drugs, drugs containing hydroalcoholic extract, and Indian ethno-medicine drugs. | (47, 48) |
| Japan (Ministry of Health, Labour and Welfare) | For over-the-counter (OTC) Kampo formulations included under “The Approval Standards for OTC Kampo products”, toxicity data and clinical studies are not required. However, toxicity data including single and repeated-dose toxicities, genotoxicity and reproductive toxicity, and clinical trial data are necessary for new Kampo formulation. | (49) |
| United States of America (FDA) | Pre-marketing approval is not required for products classified as dietary supplements, therefore safety assessment is not carried out prior to approval. However, products containing New Dietary Ingredients are subject to safety review, and botanical products used for diagnosis, alleviation of symptoms, prevention, or treatment of disease require a pre-marketing which include review of toxicological data. | (50, 51) |

**Table 2: Heavy metal and microbial contamination of natural products in Malaysia’s market**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Main plant ingredient in the product/Product type** | **Contaminant** | **Highest level detected** | **Regulatory limit (18)** | **Exceed regulatory limit**  | **References** |
| Tongkat Ali (Eurycoma longifolia)  | Mercury | 5.30 ppm | 0.5 ppm | Yes  | (52) |
| Tongkat Ali Hitam (Polyalthia bullata)  | Mercury | 2.35 ppm  | 0.5 ppm | Yes  | (53) |
| Tongkat Ali (Eurycoma longifolia) | Lead | 20.72 ppm | 10.0 ppm | Yes  | (54) |
| Itah Besi (Smilax luzonensis) | Lead | 21.21ppm | 10.0 ppm | Yes  | (55) |
| Ubi Jaga (Smilax myosotiflora) | Lead | 23.05 ppm | 10.0 ppm | Yes  | (56) |
| Kelat (Eugenia dyeriana) | Lead | 13.20 ppm  | 10.0 ppm | Yes  | (57) |
| Commercial traditional HMs  | Aflatoxin | 0.7 µg/kg | NA | NA | (58) |
| Manjakani (*Quercus infectoria*) containing post-partum products | Chromium | 4.21 ppm a | NA | NA | (59) |
| Lead | 0.17 ppm a | 10.0 ppm | No  |
| Cadmium | 0.008 a | 0.3 ppm | No  |
|  | Arsenic | 0.04 a | 5.0 ppm | No  |

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| **Table 2: (Continue)** |
| **Main plant ingredient in the product/Product type** | **Contaminant** | **Highest level detected** | **Regulatory limit (18)** | **Exceed regulatory limit**  | **References** |
| Herbal weight-loss products | Arsenic | 1.33 ppm  | 5.0 ppm | No | (60) |
| Chromium | 2.46 ppm | NA | NA |
| Zinc | 24.14 ppm | NA | NA |
| Copper | 3.54 ppm | NA | NA |
| Bacteria | 4.50 × 1011 cfu/g b  | ≤ 5 x 104 CFU/g or CFU/mL | Yes  |
| Fungi | Not enumerated. However, 16 different fungal isolates were discovered | ≤ 5 x 102 CFU/g or CFU/mL | NA |

 a Results expressed as mean concentration*;*  b The isolates were identified *Pseudomonas aeruginosa, Escherichia coli*, *Neisseria meningitidis, Staphylococcus aureus, Bacillus* spp., *Streptococcus* spp., *Enterobacteriaceae* spp., *Capnocytophaga* spp*., Corynebacterium spp., Brucella* spp., and *Yersinia* spp.; NA – Not available.

**Table 3: Advantages and limitations of alternative strategies**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Alternative strategy** | **Applications** | **Advantages** | **Limitations** | **References** |
| Chemical fingerprinting and chemometric evaluations  | * Herbal profiling
* Plant authentication
* Qualitative and quantitative assays
* Measurement of toxic contaminants
 | * Rapid and highly effective separation
* Higher selectivity and precision
* Analysis of trace levels of chemical constituents is possible
* Enables profiling of complex herbal mixtures
 | * Require advanced skills and training, and highly expensive tools and software.
 | (31, 61, 62) |

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| **Table 3: Continued** |
| **Alternative strategy** | **Applications** | **Advantages** | **Limitations** | **References** |
| Molecular docking | * Drug discovery
* CERAPP: Collaborative Estrogen Receptor Activity Prediction Project and CoMPARA: Collaborative Modelling Project for Androgen Receptor Activity
 | * Thousands of possible binding orientations can be tried and evaluated. This enables high-throughput screening of chemicals that is fast, accurate and economical
 | * Require complex computerized systems that can be expensive
* Require substantial amount of data and extensive procedures
 | (34, 75-77) |

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| **Table 3: Continued** |
| **Alternative** **strategy** | **Applications** | **Advantages** | **Limitations** | **References** |
| Omics | * Allow for potential toxicity of a drug to be predicted even in the early phases of drug discovery and development
 | * No animal sacrifice, hence fulfilling the 3Rs initiative.
* Allow study the effect of complex mixtures.
* Capable of early changes in the gene detection, such as altered cell-to-cell signalling and changes in the cell cycle, before any tumour was even formed
 | * Require ultra-low storage conditions, tedious and complicated sample preparation e.g., biological samples.
* Prone to false positive results due to enormous amount of data generated from multiple experiments.
* Require advanced training in statistical analysis, biomolecular simulation, bioinformatics, and computational skills.
 | (63-65) |

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| **Table 3: Continued** |
| **Alternative strategy** | **Applications** | **Advantages** | **Limitations** | **References** |
| (Q)SAR | * Virtual screening in early stages of drug discovery
* Screening of rodent carcinogenicity of AAI and hepatocarcinogenic estragole
* Provide additional support where existing information is inadequate, conflicting, or inconclusive to provide a rationale to support final regulatory decision (FDA)
 | * Reduce the use of animal testing
* (Q)SAR predictions fill in the data gaps when standard toxicology data are insufficient or unavailable
* Allow for interpretation of ambiguous or inadequate non-clinical study results such as genotoxicity and carcinogenicity
 | * Require complex computerized systems
* A high degree of variation in biological tests can lead to erroneous data, therefore producing inaccurate predictive models
* There is no all-in-one software to perform the modelling, which affects the learning curve and the cost of acquiring the program.
 | (33, 66-68) |

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| **Table 3: Continued** |
| **Alternative strategy** | **Applications** | **Advantages** | **Limitations** | **References** |
| PBK modelling and PBK model- based reverse dosimetry | * Personalized medicine, i.e., clinical trial design for targeted populations, e.g., elderly, paediatrics
* Derive the margin of exposure value, a harmonised approach to assess the risk of genotoxic and carcinogenic substances
 | * Allow extrapolation of data across different species, exposure routes and scenarios, and prediction for specific individuals, population, or ethnic groups
* No animal sacrifice, hence fulfilling the 3Rs initiative
* Less time, financial
 | * Only a few institutions that offer PBK modelling as a module in academic programme. As a result, there are few modelers and experts in the subject in regulatory settings
* Lack of in vivo data causes reduced availability of data for model evaluation, hence reduce the confidence of PBK model prediction
* Models are not transferable across
 | (10, 25, 32, 36, 37, 67, 69-74)  |

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| --- |
| **Table 3: Continued** |
| **Alternative strategy** | **Applications** | **Advantages** | **Limitations** | **References** |
|  | * Safety assessments for chemicals including perchlorate, acrylamide, bisphenol A and estragole
* Describe nephrotoxicity of AAI and predict developmental toxicity of all-trans-retinoic acid
 | * resources, and workforce required.
 | * different modelling software
 |  |

Natural products

Drug Control Authority (DCA)

National Pharmaceutical Regulatory Agency
(NPRA)

Drug Registration Guidance Document (DRGD)

Post-Market Activities

Pre-Market Safety Assessment

1. Post-Market surveillance - Registered products may be sampled and tested
2. Product complaints
3. Pharmacovigilance – ADR reporting
4. Evidence on safety – e.g., Scientific reports, articles /bibliographic evidence/monographs. Toxicological data and clinical studies compulsory for natural products with disease reduction claim
5. Free from prohibited/banned substances
6. Quality evidence – e.g., quality control documents, certificate of analysis (COA) on active ingredients and COA on finished products, limit test for heavy metals, disintegration and weight uniformity tests for tablets, capsules, and pills, and test for microbial contamination
7. Stability data – real-time and accelerated
8. GMP compliance certificate

**Fig 1:** **Overview of the registration process for natural products in Malaysia. All natural products must be registered with Drug Control Authority (DCA). National Pharmaceutical Regulatory Agency (NPRA) which acts as the secretariat, published the Drug Registration Guidance Document (DRGD). It contains detailed guidance pertaining registration process and post-registration activities of medicinal products.**

**Fig 2: Adverse drug reactions (ADR) reports attributed to natural products in Malaysia (78)**