BMB Reports - Manuscript Submission

Manuscript Draft

Manuscript Number: BMB-22-102

Title: Systems pharmacology approaches in herbal medicine research: A brief review

Article Type: Mini Review

Keywords: Herbal medicine; traditional medicine; systems pharmacology; network pharmacology; drug-induced transcriptome

Corresponding Author: Haeseung Lee

Authors: Myunggyo Lee[1], Hyejin Shin[1], Musun Park[1], Aeyung Kim[1], Seongwon Cha[1], Haeseung Lee[1,*]

Institution: [1]College of Pharmacy, Pusan National University, Busan, 46241, Korea,
[2]Korean Medicine (KM) Convergence Research Division and [3]Korean Medicine (KM) Data Division, Korea Institute of Oriental Medicine, Daejeon 34054, Republic of Korea,
[4]Korean Medicine (KM) Application Center, Korea Institute of Oriental Medicine, Daegu 41062, Republic of Korea,

Manuscript Type: Mini Review

Title: Systems pharmacology approaches in herbal medicine research: A brief review

Author's name:

Myunggyo Lee[1,*], Hyejin Shin[2,*], Musun Park[3], Aeyung Kim[4], Seongwon Cha[3], Haeseung Lee[1]

Affiliation:

[1] College of Pharmacy, Pusan National University, Busan, 46241, Korea

[2] Korean Medicine (KM) Convergence Research Division, Korea Institute of Oriental Medicine, Daejeon 34054, Republic of Korea.

[3] Korean Medicine (KM) Data Division, Korea Institute of Oriental Medicine, Daejeon 34054, Republic of Korea.

[4] Korean Medicine (KM) Application Center, Korea Institute of Oriental Medicine, Daegu 41062, Republic of Korea

* These authors contributed equally to this work.

Running Title: Systems pharmacology for herbal medicine research

Keywords: Herbal medicine, traditional medicine, systems pharmacology, network pharmacology, drug-induced transcriptome

Corresponding Author's Information:

Haeseung Lee

Tel. No.: +82-51-510-2823; E-mail: haeseung@pusan.ac.kr

ABSTRACT

Herbal medicine, a multi-component treatment, has been extensively practiced for treating various symptoms and diseases. However, its molecular mechanism of action on the human body is unknown, which impedes the development and application of herbal medicine. To address this, recent studies are increasingly adopting systems pharmacology, which interprets pharmacological effects of drugs from consequences of the interaction networks that drugs might have. Most conventional network-based approaches collect associations of herb-compound, compound-target, and target-disease from individual databases, respectively, and construct an integrated network of herb-compound-target-disease to study the complex mechanisms underlying herbal treatment. More recently, rapid advances in high-throughput omics technology have led numerous studies to exploring gene expression profiles induced by herbal treatments to elicit information on direct associations between herbs and genes at the genome-wide scale. In this review, we summarize key databases and computational methods utilized in systems pharmacology for studying herbal medicine. We also highlight recent studies that identify modes of action or novel indications of herbal medicine by harnessing drug-induced transcriptome data.

Current trends in systems pharmacology for herbal medicine research

Medicinal herbs have been used extensively for the treatment of various ailments in ancient medical traditions of China (traditional Chinese medicine, TCM), Korea (traditional Korean medicine), Japan (Kampo medicine), India (Ayurveda), Indonesia (Jamu), North America (phytotherapy), and Europe (herbalism) (1). Standardized herbal extracts (hereafter referred to as herbs) or herbal formulae that blend several herbs into a single formula are composed of a variety of bioactive chemical compounds. They provide a fertile ground for modern drug development with therapeutic leads. Antimalarial quinine and artemisinin, antipyretic analgesic aspirin, and arsenic trioxide for leukemia are examples of modern drugs originally used in traditional medicine (2). In general, pharmacological effects of herbal medicine are achieved by their active ingredients that simultaneously modulate multiple biomolecules in the human body via an additive or synergistic manner. This multi-component nature of herbal medicine has been considered advantageous over single-target drugs for treating complex multifactorial disorders such as cancer and nervous system disease (3, 4). Although therapeutic effects of herbal medicine have been clinically verified in traditional settings for thousands of years, its unknown mode of action on the human body hinders its application and development.

Along with great progress in systems biology, systems pharmacology or network pharmacology approaches have been introduced to decipher complex mechanisms of action (MOAs) of drugs in networks of biomolecules that interact with the drugs (5-7). These approaches have been extensively applied to explore pharmacological effects of multi-component herbal medicine acting on multiple targets of disease from a holistic perspective (8). A typical network pharmacology approach starts from constructing a network in which a node represents either an herb, herbal ingredient, or target protein/gene and an edge indicates link between herb and its constituent compound or interaction between compound and its target

(Figure 1). By associating targets with biological functions or diseases, an herb-compound-target-function/disease network is constructed and leveraged to study the MOA of an herbal treatment. Protein-protein interaction (PPI) network could be further employed to interpret the synergistic effect of herbal medicine by analyzing interactions among target proteins (9, 10). As this network-based approach is solely based on the network constructed, a significant factor influencing subsequent analysis is the reliability of connections (herb-compound, compound-target, and target-disease) in the network. However, establishing reliable connections requires laborious tasks such as identifying herbal ingredients and their molecular targets (11). Thus, most network pharmacology studies only cover a fraction of herbal compounds quantified and rely on target annotations based on *in silico* prediction. Moreover, although effects of compounds on targets depend on biological systems perturbed (e.g., tissue or cell), most of the available information on compound-target interactions have been obtained by high-content screenings in cell-free systems. In addition, even if reliable associations between compounds and targets are available, it is difficult to identify downstream genes affected by the targets to exert a therapeutic effect.

One emerging alternative to address these issues is leveraging drug-induced transcriptome data. The gene expression profile induced by herbal treatment reflects genome-wide effects of multi-component herbs in a certain biological system, thereby providing comprehensive and reliable associations between herbs and genes (12) (Figure 1). A typical application of drug-induced transcriptome data was first introduced systematically by the connectivity map (CMap) (13). CMap currently provides large-scale gene expression profiles before and after treatment with ~33,000 small molecule compounds in 230 human cell lines and periodically releases additional data (14). It has been widely utilized in research to retrieve drug repositioning candidates and to elucidate drug's MOA in modern and herbal medicine (15, 16). With a similar concept, drug-induced transcriptome data from MCF-7 breast cancer cell

line treated with each of 102 TCM components (TCM102) have been published (17), enabling researchers to explore the activity of TCM ingredients at the molecular and cellular levels (18, 19). As more and more herbal medicine studies conducting high-throughput transcriptome profiling have been published, numbers of gene expression data sets of herbs/ingredients generated in various experimental settings have been accumulated. To integrate these data, Fang S and colleagues have collected transcriptome data sets of 20 herbs and 152 ingredients and built an organized database, HERB, a high-throughput experiment- and reference-guided database of TCM (20). As demonstrated by the increasing demand for these key resources, analyzing transcriptome data of herbal treatment can efficiently uncover novel associations between herbal medicine and modern drugs, genes, and diseases, which in turn can encourage the application and development of herbal medicine.

Systems pharmacology approaches using network-based methods or drug-induced transcriptome data are increasingly adopted pivotally in herbal medicine research. In this review, we summarize key databases and computational methods used in systems pharmacology for studying herbal medicine. We also highlight the recent application of pharmaco-transcriptomics in herbal medicine research.

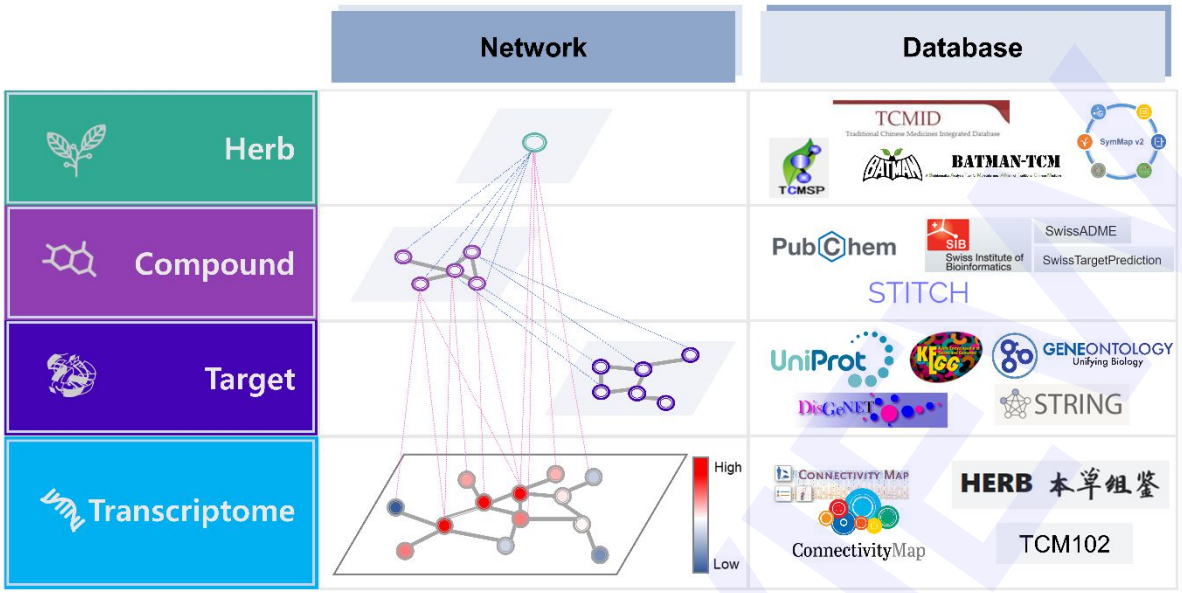

Figure 1. Resources for network pharmacology in herbal medicine research

Left shows typical forms of an herb-compound-target network (blue edges) and herb-gene or compound-gene network (pink edges) used in network pharmacology research. Right shows public databases frequently utilized to construct networks for herbal medicine research.

Public databases used in systems pharmacology for herbal medicine research

Systems pharmacology approaches for herbal medicine usually start with integrating current knowledge on different types of data (including herbs, compounds, targets, pathways, and diseases) and organizing them into a network. Typically, in the network, a node represents an herb, compound, target, pathway, or disease and an edge represents an interaction between nodes. Numerous databases provide information on the nodes or edges, each database containing data of different scopes and evidence levels. These databases can be divided into four types: herb-related (HRDB), compound-related (CRDB), target-related (TRDB), and disease-related (DRDB) databases (Table 1). Information on herbal properties and herbal ingredients can be obtained from HRDBs, such as TCMID (21), TCMSP (22), BATMAN-TCM (23), and SYMMAP (24). Most HRDBs provide not only information on herbal ingredients,

but also ingredient-related targets, pathways, and diseases. For this reason, HRDBs have been mainly utilized for network pharmacology analysis of herbal medicine. The CRDB includes databases for compounds, compound-target interaction (CTI), and compound-induced transcriptome data. PubChem (25) and SwissADME (26) contain information on physicochemical descriptors, pharmacokinetic properties, and ADME (absorption, distribution, metabolism, and excretion) parameters of compounds, which can be used to calculate drug-likeness values of herbal compounds. SwissTargetPrediction (27), STITCH (28), and Therapeutic Target Database (TTD) (29) provide known or predicted CTI information. Although high-throughput targeted assays have been developed to screen for drug targets, it is a fairly arduous task to identify binding targets on a genome-wide scale, in some cases for hundreds of herbal ingredients. Therefore, some CRDBs additionally provide information on CTIs predicted by using machine learning (30) or similarity (31) based on structures of compounds and targets. Among TRDBs, Uniprot (32) and GeneCards (33) provide information on the sequences and functional roles of proteins/genes. KEGG (34), Reactome (35), and gene ontology (36) provide sets of genes classified by their biological functions. These databases are utilized to perform functional enrichment analysis, such as GSEA (gene set enrichment analysis) (37). In addition, STRING (38) and Human Protein Reference Database (39) provide known PPI information. PPI information can be integrated with CTI to construct a target network in which potential drug targets are identified as interacted protein modules in the network (40). DRDB provides collections of genes and variants associated with diseases. DRDB is also widely used because one of the ultimate goals of systems pharmacology is to predict and evaluate therapeutic effects of drugs by exploring the relationship between drugs and diseases.

Table 1. Public databases widely used in herbal medicine research

Type	Database	Numbers of available data	Website or Reference
Herb-related database	TCMID (Version 2.0)	46,929 TAM prescriptions 8,159 herbs / 43,413 compounds	http://www.megabionet.org/tcmid/
	TCMSP (Version 2.3)	501 herbs / 13,144 compounds 3,311 targets / 837 diseases 29,384 HC pairs / 84,260 CT pairs 2,387 TD pairs	https://old.tcmsp-e.com/tcmsp.php
	SYMMAP (Version 2.0)	698 herbs / 26,035 compounds 20,965 targets /14,086 diseases 2,518 TAM symptoms 1,148 MM symptoms	http://www.symmap.org/
Compound-related database	PubChem (2021)	111 million compounds 278 million substances 295 million bioactivities	https://pubchem.ncbi.nlm.nih.gov/
	STITCH (v5.0)	430,000 compounds 9,600,000 proteins	http://stitch.embl.de/
	CMap	33,000 compounds / 230 cell lines	https://clue.io/
	TCM102	102 compounds 1 cell line (MCF-7)	(17)
	HERB	7,263 herbs / 28,212 compounds 12933 targets / 49,258 phenotypes 6,164 gene expression profiles	http://herb.ac.cn/
Target-related database	UniProt (2020.04)	292,000 proteins (190 million sequences)	https://www.uniprot.org/
	KEGG (2022.03.24)	551 biological pathways	https://www.genome.jp/kegg/
	Gene Ontology (2022.03.22.)	7,838,790 gene sets involved in biological process, molecular function, and cellular components	http://geneontology.org/
	STRING (v11.5)	24,584,628 proteins 5,090 organisms	https://string-db.org/
Disease-	DisGeNet	21,671 genes / 30,170 diseases	https://www.disgenet

related database	(v7.0)	1,134,942 gene-disease associations	.org/ (41)
	OMIM (2022.05.02)	16,730 genes / 6,378 phenotypes	https://www.omim.org/ (42)
	Human Phenotype Ontology (2022.04)	4,791 genes / 10,274 phenotypes	https://hpo.jax.org (43)

TAM, traditional Asian medicine; MM, modern medicine.

Computational approaches for studying herbal medicine

With the introduction of polypharmacology, the paradigm of drug research has shifted from single-target to multi-target strategies, revealing the potential of multicomponent herbal medicines to treat a variety of multifactorial disorders (44-46). In line with this, various computational approaches have been introduced to identify targets, indications, and/or synergistic combinations of herbal medicines (Table 2).

Prediction of pharmacological targets

Network-based methods have been most widely applied to predict potential targets of herbs or herbal formulae. For example, Wang et al. (47) have predicted synergistic targets of four herbs, *Radix Astragali Mongolici* (RAM), *Radix Puerariae Lobatae* (RPL), *Radix Ophiopogonis Japonici* (ROJ), and *Radix Salviae Miltiorrhiza* (RSM) in cardiovascular diseases (CVD). They selected bioactive compounds of the four herbs by considering compounds' drug-likeness and oral bioavailability. Targets of the compounds were then predicted based on their structural and physicochemical properties via a machine learning method (30). Finally, synergistic effects of RSM and other herbs were interpreted from target

proteins shared by herbs in the herb-compound-target network. For another example, Li et al. (48) have investigated pharmacological targets of an herbal formula Qishenkeli (QSKL) by analyzing QSKL-induced transcriptome data of myocardial ischemia pig model. QSKL is a traditional herbal mixture used for routine treatment of CVD in China. It consists of six herbs: *Astragalus propinquus* Schischkin root and rhizome, *Salvia miltiorrhiza* Bunge root and rhizome, *Lonicera japonica* Thunb flower, *Aconitum carmichaelii* Debeaux lateral root, *Glycyrrhiza glabra L.* root and rhizome, and *Scrophularia ningpoensis* Hemsl root. Based on the hypothesis that drugs with similar drug-induced expression patterns share the same MOA, they virtually screened drugs with expression patterns similar to QSKL from CMap database. Known targets of these drugs were collected from DrugBank and considered as potential pharmacological targets of QSKL. As a result, MOA of QSKL was interpreted from a QSKL-drug-target network that included 18 drugs within the ATC (anatomical therapeutic chemical) cardiovascular system category and their known target genes such as CPT-1 and CPT-2 involved in cardiac energy metabolism.

Several machine learning methods have also been employed to predict herb-target interactions (49-51). Wang et al. (49) have presented a network integration pipeline HTINet (Herb-Target Interaction Network) for herb-target prediction. HTINet firstly constructs a heterogeneous network by integrating five types of nodes (herb, target, drug, disease, symptom) and edges as their corresponding interactions from diverse data sets. From the network, feature representations of herb and protein nodes are extracted using a network embedding algorithm called node2vec (52). Finally, a classification model to predict herb-target interactions is built by applying a supervised learning method K-Nearest Neighbor (KNN), which learns rules from the known relationships between herbs and targets based on their features. In a similar way, Zhao et al. (50) have proposed a herb-target prediction method called HGNA-HTI (Heterogeneous Graph Neural Network with Attention Mechanism for Prediction of Herb-

Target Interactions), which has three parts: i) constructing a heterogeneous herb-target network from diverse data sets, ii) learning feature representations of herbs and targets using an attention mechanism with respect to the topological structure of the network, and iii) predicting interactions between herbs and targets by link prediction based on features. As another example, Keum et al. (51) have employed classification models that predict the interactions between targets and herbal compounds by utilizing information on approved drugs, target proteins, and known interactions thereof. They used chemical structural similarities of drugs and sequence similarities of proteins as feature representations of drugs and targets, respectively. Similarities of drugs and proteins were calculated using the Open Babel fingerprint and the Smith-Waterman algorithm, respectively. Using these features, a bipartite local model (53) was trained for predicting drug-target associations. Prediction models were generated separately depending on functional classes of target proteins, including G-protein-coupled receptors (GPCRs), enzymes, transporters, receptors, and other proteins. A list of herbal compounds was obtained from TCMID, TCM-ID, KTKP, and KAMPO. Chemical structure information was taken from ChemSpider. Lastly, target proteins of herbal compounds were predicted by applying the constructed classification models.

Prediction of pharmacological indications

Active compounds stemming from medicinal herbs are appealing in modern drug development due to their high efficacies and low toxicities (2, 54). However, new therapeutic opportunities for numerous herbal compounds are yet to be identified. In this section, we will review a few studies applying state-of-the-art machine learning methods to prioritize therapeutically effective herbal compounds for several diseases (55-58).

Yoo *et al*. (55) have introduced a network-based method for identifying pharmacological effects of herbal compounds based on phenotype-related associations. They

applied a random walk with restart (RWR) algorithm to generate phenotype vectors of herbs based on associations between known efficacies of herbs and thousands of phenotypes in a phenotypic network. The phenotypic network represents the hierarchical relationship of the Unified Medical Language System (UMLS) which provides integrated information for various phenotypic terms. Hierarchical clustering of phenotype vectors was conducted to extract herb clusters with similar efficacies. In an herb cluster, significantly enriched herbal compounds were selected using Fisher's exact test. These compounds were considered to have pharmacological effects that the herb cluster had. The same group further applied a deep neural network approach to predict medicinal uses of herbal compounds based on molecular and chemical features of approved drugs (56). They extracted feature representations of approved drugs and herbal compounds using three types of data: i) latent knowledge of drugs obtained by text mining of scientific literature, ii) molecular interactions of drugs in the PPI network, and iii) chemical properties, including physicochemical properties, lipophilicity, pharmacokinetics, and drug-likeness. A deep learning model was then trained based on extracted features and verified indication information of drugs. Using the trained model, potential medicinal uses of herbal compounds were predicted based on extracted features of compounds. As another example, Kim et al. (58) have applied several classification algorithms to predict new indications for herbal compounds. They hypothesized that similar drugs could treat similar diseases. They used similarities for both drug and disease aspects as predictive features to predict novel drug-disease associations. Drug-drug similarity was calculated using four types of data (chemical structure, side-effects, gene ontology, and targets) and disease-disease similarity was calculated using three types of data (phenotypes, human phenotype ontology, and gene ontology). As a training data set, information on drugs, diseases, and drug-disease associations were obtained from DrugBank, Online Mendelian Inheritance in Man (OMIM), and a previous study (59), respectively. Prediction models were constructed using

three machine learning algorithms: logistic regression, random forest, and support vector machine algorithms. They showed that the random forest approach achieved the best performance upon cross-validation and external validation using independent dataset (58).

Prediction of synergistic combinations

Synergy is one of the major advantages of multi-component herbal medicine. Network-based strategies enable us to efficiently explore herb combinations and to better infer the mechanisms of synergistic action of herbs or herbal compounds. For example, Li et al. (60) have proposed an effective herbal combination for CVD from CVD-associated compound-target networks. They collected information on known targets related to CVD from DrugBank, TTD, and a previous study (61) and predicted their potential ligands via a machine learning method (30). Based on this prediction, a bipartite network of compound-target was built. To find effective herbs for CVD, ligand compounds in the network were mapped to herbs that contained corresponding compounds using TCMSP database. Considering that herbal formulae consist of multiple herbs with different roles (62), *Radix Salviae Miltiorrhizae* (RSM) was first selected with expectation of its dominant role in CVD treatment as it was associated with many CVD-related targets and pathways. Li et al. (60) have hypothesized that if two herbs affect the same functional pathways, they would have pharmacological synergy against a disease. Therefore, additional herbs acting on the same pathways as those of RSM were recruited as follows: *Carthamus tinctorius* (CT) and *Fructus Cartaegi* (FC). Finally, a mixture of RSM, CT, and FC was proposed as a novel herb combination for the treatment of CVD. Therapeutic effects of the mixture were further validated in a mouse model of myocardial infarction.

As an another example, Wang *et al*. (63) have proposed a network-based method to infer the molecular mechanisms of synergistic interactions of herbs. Considering that traditional herbal formulae were completed based on the principle of herb compatibility, they

assumed that herb pairs in the existing herbal formulae would be more synergistic than not in the herbal formulae. To examine this assumption, they developed a network proximity model quantifying the degree of synergistic herb interactions. Firstly, information on herbal ingredients and their targets related to all herb pairs within herbal formulae were collected from TCMID and STITCH, respectively. This information was integrated into the human PPI network, resulting in a network consisting of three levels of interactions, including herb-ingredient, ingredient-target, and target-target for each herb pair. Wang et al. then measured the network proximity of each set of targets of a given herb pair in the PPI network and showed that commonly used herb pairs tended to affect proteins at shorter distances in the PPI network compared to random herb pairs. In addition, they found that ingredients located at the center of the herb PPI network played an important role in their synergy with other herbs. It implies that this network proximity model is feasible to prioritize active ingredients producing combinational effects of herb pairs.

Table 2. Computational approaches for studying herbal medicine

Reference	**Prediction Type**	***in silico* Tools Utilized**	**Data Sources Utilized**
Wang *et al*., 2012 (47)	herb-target interactions	ligand-target interaction prediction (30)	TCMSP (22), DrugBank (64), PharmGKB (65), TTD (29)
Li *et al*., 2017 (48)	herb-target interactions	ligand-target interaction prediction (30)	TCMSP, DrugBank, CMap
Wang *et al*., 2019 (49)	herb-target interactions	node2vec, KNN, SVM, RF, LR, DT, GBDT	HIT (66), Chinese pharmacopoeia, SIDER (67), MalaCards (68), DrugBank, SemMedDB (69), Zhou *et al*. (70), STRING (38)
Zhao *et al*., 2021 (50)	herb-target interactions	GNN (71), meta relation-based attention mechanism	HeNetRW (72), YaTCM (73), TCMIP (74)
Keum *et al*., 2016 (51)	herb-target interactions	BLM(53), SVM	DrugBank, TCMID (21), TCM-ID (75), KTKP (http://www.koreantk.com), KAMPO (http://kampo.ca/),

			ChemSpider (http://www.chemspider.com/)
Yoo *et al*., 2018 (55)	indications of herbal compounds	RWR, hierarchical clustering	OMIM (42), KTKP, MeSH, TCMID, TCMSP, TCM@Taiwan (76), TCM-ID, KAMPO
Yoo *et al*., 2020 (56)	indications of herbal compounds	RWR, DNN	MeSH, OMIM, KTKP, TCMID, COCONUT (77), FooDB (http://foodb.ca/), DrugBank, CTD (78), MATADOR (79), STITCH (28), TTD, BioGrid (80)
Kim *et al*., 2019 (58)	indications of herbal compounds	LR, RF, SVM	DrugBank, OMIM, SIDER, OFFSIDES (81), STITCH, UniProt (82), DGIdb (83), HPO (84), DisGeNet (85), KTKP, TCMID, TCM-ID, KAMPO, BindingDB (86), MATADOR
Li *et al*., 2014 (60)	effective combination of herbs	ligand-target interaction prediction (30)	DrugBank, TTD, TCMSP
Wang *et al*., 2021 (63)	synergistic MOA of herbs	network proximity measure (87)	TCMID, STITCH, Cheng *et al*. (87)

KNN, K-Nearest Neighbor; SVM, support vector machine; RF, Random forest; LR, Logistic Regression; DT, Decision Tree; GBDT, Gradient Boosting Decision Tree; GNN, Graph Neural Network; BLM, Bipartite Local Model; RWR, Random walk with restart; DNN, Deep Neural Network; GTB, Gradient Tree Boosting; OMIM, Online Mendelian Inheritance in Man; MOA, mechanism of action

Transcriptome-based elucidation of molecular target and/or MOA of herbal medicine

Herbal prescriptions are combinations of herbal formulas for treatment, meaning that various ingredients in these formulas have the potential to affect multiple genes and biological pathways. In recent decades, systems pharmacology studies applying transcriptome analysis after treatment with herbal medicines *in vitro* or *in vivo* have elucidated the mechanisms of herbal effect in various diseases (88). In this section, we will look at several studies investigating the molecular targets and MOA of herbal medicines using transcriptome data.

Si-Wu-Tang (SWT) (Samul-tang in Korean, Shimotsu-to in Japanese) is one of the most popular herbal prescriptions consisting of four herbs including *Paeonia Radix, Ligusticum Rhizoma, Rehmannia Radix,* and *Angelica Radix* at a 1:1:1:1 ratio. It has been clinically applied for centuries in Asia to treat symptoms of hematological disorders such as anemia, menstrual irregularities, dysmenorrhea, and menopausal syndrome (89-97). In recent years, an increasing number of studies have elucidated the MOA of SWT by analyzing SWT-induced changes in gene expression. Wen et al. (91) have reported the MOA and therapeutic evidence of SWT using SWT-induced transcriptome data of MCF-7 cells and CMap data for the first time. Gene expression profiles of SWT-treated MCF-7 cells were significantly matched with those of estradiol-treated MCF-7 cells in the CMap database, suggesting that SWT might have potential phytoestrogenic effects. Pathway enrichment analysis using differentially expressed genes (DEGs) of SWT-treated datasets revealed that *Nrf2*-regulated genes, antioxidant genes, and chemopreventive inducible genes were affected by SWT treatment, but not by estradiol or ferulic acid in MCF-7 cells. The potential role for phytoestrogen of SWT was further confirmed by whole genome microarray profiling and by estrogen-responsive element luciferase reporter assay in MCF-7 cells (92), suggesting the potential of SWT as an estrogen receptor modulator. Another *in silico* analysis (93) predicted SWT's new targets, FOS, JUN, and CAPS3 based on SWT-induced transcriptome data and herb-target information obtained from TCMID database.

These studies provide insights into the understanding of complex actions of SWT for gynecological diseases. Antioxidative and estradiol-regulating effects of SWT may help ameliorate ovarian follicular maldevelopment associated with ovulatory and menstrual disorders known to cause infertility (94). Recently, protective effects of SWT on ovarian function and oocyte maturation against cyclophosphamide (CP, a chemotherapy drug)-induced chronic ovarian dysfunction and maternal aging have been demonstrated using transcriptome analysis in an *in vivo* mouse model (95, 96). DEGs by SWT treatment in CP-treated mice ovaries were involved in ovarian follicle development, binding of sperm to zona pellucida (Zp), and microtubule nucleation (95). Among downregulated DEGs, oocyte-specific genes such as fertilization-related genes (*Zp2*, *Zp3*, *Nlrp5*), ovarian follicle development-related genes (*Bmp15*, *Oas1d*), and oocyte maturation-related genes (*Obox1*) were restored to normal levels after SWT administration. Additionally, further enrichment analysis using TargetScan, a tool for sequence-based microRNA-target predictions, identified microRNA binding sites of DEGs. Three microRNAs (miR-200b-3p, miR-665-3p, and miR-667-3p) had binding sites on either *Bmp15* or *Oas1* mRNA and their expression levels were restored in SWT plus CP-treated mice. Kim and You have also investigated the influence of SWT on ovarian reserve and fertilization in aged mice (> 40-week-old) (96, 97). They performed functional enrichment analysis of transcriptome data obtained from ovulated ovaries and revealed that RAS signaling pathway-related genes were restored to normal levels as those in young mice after SWT administration to aged mice (96). SWT administration also restored miR-223-3p, which could interact with embryo implantation-related genes (*Hal, Acp5*) in the uterus of aged mice (97). This transcriptome analysis result was consistent with the phenotypic findings of the protective effect of SWT on the increase of ovarian reserve, ovulated mature oocytes, and pregnancy rate in aged mice (96, 97).

Tao-Hong-Si-Wu Decoction (THSWD), a traditional herbal medicine, is composed of

six herbs (*Prunus persica*, *Carthamus tinctorius*, *Rehmannia glutinosa*, *Angelica sinensis*, *Paeonia lactiflora*, and *Ligustium chuanxiong*). It has been used for clinical treatment of gynecological and cerebrovascular diseases (98, 99). Duan et al. (99) have performed transcriptome profiling of a rat stroke model treated with THSWD and functional enrichment analysis of DEGs induced by THSWD. From enriched functional terms, cell cycle, complement and coagulation cascades, and neuroactive ligand–receptor interactions were suggested as potential therapeutic targets of THSWD for intracerebral hemorrhage-induced neurological deficits.

Compound Kushen Injection (CKI) is an approved Chinese patented drug in adjuvant treatment for chemotherapy. It consists of extracts of two herbs, Kushen (*Sophora flavescens*) and Baituling (*Heterosmilax chinensis*). Qu et al. (100) have leveraged CKI-induced transcriptome data of MCF-7 cells and revealed that CKI primarily downregulates most genes functioning in the cell cycle in a similar degree to a chemotherapeutical agent, 5-fluorouracil (5-FU). Interestingly, although CKI exerted anticancer effects on MCF-7 cells, the expression of *TP53,* a pro-apoptotic gene, was decreased by CKI treatment but increased by 5-FU. From this observation, they proposed that CKI might induce MCF-7 cell apoptosis via a p53 independent pathway. The anticancer effect of CKI was also confirmed via transcriptome analysis using single herb extracts in another breast cancer cell model, MDA-MB-231 (101). CKI has complementary effects on cancer cells as Kushen perturbs cell cycle regulation whereas Baituling activates the immune system. Further transcriptome analysis and cell migration assay revealed that CKI could control cancer metastasis in multiple cancer cell lines of colon (HT-29, SW-480, DLD-1), brain (U87-MG, U251-MG), and breast (MDA-MB-231) (102). To identify core CKI response genes in cancer cells, further investigation was conducted to compare MCF-7 CKI-DEGs to Hep G2 (liver cancer) CKI-DEGs and MDA-MB-231 CKI-DEGs by Cui et al. (103). They found that eight components of CKI obtained from BATMAN

tools potentially interacted with 52 core genes in integrated DEGs in multiple cancer cell lines.

Feifukang (FFK) is a pulmonary rehabilitation mixture comprising eight herbs for protecting lung function: *Astragalus membranaceus*, *Codonopsis pilosula*, *Ophiopogon japonicus*, *Schisandra chinensis*, *Panax notoginseng*, *Bulbus fritillariae thunbergii*, *Rhizoma anemarrhenae*, and *Glycyrrhiza uralensis* (104, 105). Li et al. (105) have evaluated anti-pulmonary fibrosis effect of FFK in a bleomycin (BLM)-induced pulmonary fibrosis mouse model and explored targets of FFK by analyzing transcriptome data of FFK-treated mice. FFK has potential antifibrotic effects by significantly reducing collagen fiber formation in BLM-induced lung fibrosis. Functional enrichment analysis of DEGs in FFK-treated mice showed that the JAK-STAT signaling pathway was significantly downregulated by FFK. qRT-PCR and western blot analysis of *JAK1*, *STAT3*, and *ADAM17* as representative genes confirmed the effect of FFK in regulating lung fibrosis through JAK-STAT signaling pathway.

Paeoniae Radix (PR), a root of the plant *Paeonia lactiflora*, is a key material in many herbal treatments. It is known to supply blood, prevent sweating, regulate menstruation, and relieve pain (106). Baek et al. (107) have identified a novel anticancer mechanism and activity of PR by analyzing transcriptome data derived from lung cancer cells treated with PR extracts or its ingredients. They performed a series of GSEA based on dose-dependent PR-induced transcriptome profiles of lung cancer cells and showed that PR and its two ingredients, hederagenin and oleanolic acid, exerted anticancer effects on lung cancer by downregulating the Aurora B pathway. The synergistic MOA of PR was further interpreted via an integrated PR extracts-compounds-target genes network in the Aurora B pathway.

These individual studies on specific herbal formulae or an herb have laid the basis for developing efficient strategies to systematically infer MOA of herbal medicines at the molecular level, which may rationalize and modernize herbal medicines ultimately.

Transcriptome-based identification of novel indications for herbal compounds

Another great advantage of obtaining drug-induced transcriptome data of herbs/ingredients is that novel indications of herbal medicine can be rapidly screened computationally by a systems-based approach. A systems-based approach involves modulating a list of abnormally expressed genes in disease, in contrast to a traditional target-based approach which involves modulating the molecular state of one single protein (108). This approach was first designed and introduced to the public by CMap to link drugs and diseases (13). It defines a set of abnormally expressed genes in a disease, termed a disease signature, and queries it in the CMap reference database. It then searches for drugs that inversely regulate the expression of the disease signature, that is, those that decrease the expression of upregulated disease genes and increase the expression of downregulated disease genes. These drugs are considered candidates for reversing the diseased state back to the normal state.

Several studies using this approach have demonstrated its applicability to drug repositioning of herbal compounds (109, 110). Luo et al. (109) have identified flavonoid luteolin as a therapeutic agent for ischemic stroke (IS) via inhibiting *MMP9* and activating PI3K/Akt signaling pathways. Flavonoids are common constituents of plants used in traditional herbal medicine to treat a wide range of diseases (111). Luteolin was predicted by CMap analysis finding compounds that could regulate the expression of IS signature obtained from transcriptome data of IS patients. Further in vivo experiments demonstrated that luteolin reduced the infarct volume in a rat model of IS. Similarly, Liu et al. (110) have utilized the CMap database to seek compounds that could mimic transcriptional changes induced by a variety of interventions to reduce endoplasmic reticulum (ER) stress based on the observation that increased ER stress could develop leptin resistance and lead to obesity. As a result, they identified celastrol, a pentacyclic triterpene extracted from roots of *Thunder of God Vine* plant

as a promising agent for treating obesity by increasing leptin sensitivity. They showed that celastrol could reduce ER stress and increase leptin sensitivity to reduce body weights of obese mice.

TCM102 database is also widely utilized in research to discover new indications of herbal compounds based on the systems-based approach (18, 19). For example, Li et al. (18) have identified several vasodilators from TCM102 database. They first generated two gene signatures that involved in positive and negative regulation of blood vessel diameter by using databases GO and SEEK (search-based exploration of expression compendia) (112). Compounds that inversely regulated the expression of these two gene signatures were then screened by performing GSEA on drug-induced transcriptome data in TCM102. Top 10 candidate compounds were tested for their vasorelaxant effects on vascular tension of constricted thoracic aortic rings in a rat model. Among these 10 compounds, ferulic acid exhibited the strongest vasorelaxant effect, and others also induced relaxation significantly. Finally, mechanisms of six compounds (ferulic acid, borneol, daidzin, magnolol, chenodeoxycholic acid, and artemisinin) were inferred from one integrated network representing pathways involving signature genes specifically regulated by these six compounds. As another example, Wang et al. (19) have utilized TCM102 database for the evaluation of phillyrin as an anti-cardiac fibrosis agent. Phillyrin was of interest because it was one of the major active ingredients of *Forsythia suspensa*, an herbal medicine used as an anti-inflammatory and antipyretic drug (113). They first analyzed transcriptome data sets of human heart diseases and constructed a cardiac fibrosis (CF)-related gene functional module (CFGM), which had a set of genes containing three known CF markers, *Postn*, *Ddr2*, and *Pdgfra,* and their co-expressed genes. They found that treatment with phillyrin reduced expression levels of most CFGM members from phillyrin-induced transcriptome data in TCM102. They hypothesized that drugs decreasing the expression of CFGM had the potential to treat CF by

inhibiting the core pathological process of CF. Cardio-protective and anti-CF effects of phillyrin were further validated in a myocardial infarction rat model.

These systems-based approaches have been mainly conducted by screening desired compounds using well-organized databases, such as CMap and TCM102. However, since these databases only contain data on small molecule compounds, herbs or herbal formulae are inevitably excluded from the screening. The expanded database including a variety of medicinal herbs would offer clues to identify evidence-based connections between herbs and diseases, hence spurring the application and development of herbal medicines.

CLOSING REMARKS

Systems pharmacology approach is increasingly adopted and developed in a wide range of modern drug development processes to better understand molecular MOA of drugs in the human body. Although this approach would also lend itself to herbal medicine research, its practical application and development are relatively slow. The main reason is that the data on herbs and herbal medicines themselves are insufficient for systems pharmacology approach to directly utilize. Whereas for modern drugs, CMap alone provides drug-induced transcriptome data for ~40,000 small molecules, and furthermore, the Library of Integrated Network-Based Cellular Signatures (LINCS) project is continuously generating drug-related multi-omics data sets including proteome, epigenome, and metabolome data for a comprehensive understanding of drug MOA. The advantage of such large-scale data is that we can rapidly utilize them to repurpose existing drugs in urgent situations. For example, several approved drugs have been proposed as candidates for clinical intervention to combat rapidly emerging diseases such as COVID-19 through *in silico* screening using CMap data (114, 115). Accordingly, the establishment of a well-organized drug-related database (e.g., drug-induced transcriptome data) for standardized herbs or herbal medicines should pave the way for advancement of herbal

medicine research.

ACKNOWLEDGMENTS

This work has been supported by the National Research Foundation of Korea (NRF) grant funded by the Korea government (MSIT) (NRF-2021R1C1C1003988) and the research program of the Korea Institute of Oriental Medicine (KIOM) (KSN2023120 and KSN2022240).

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

FIGURE LEGENDS

Figure 1. Resources for network pharmacology in herbal medicine research

Table 1. Public databases widely used in herbal medicine research

Table 2. Computational approaches for studying herbal medicine

REFERENCES

1. Zhao X, Zheng X, Fan T-P, Li Z, Zhang Y and Zheng J (2015) A novel drug discovery strategy inspired by traditional medicine philosophies. Science 347, S38-S40
2. Corson TW and Crews CM (2007) Molecular understanding and modern application of traditional medicines: triumphs and trials. Cell 130, 769-774
3. Stancak B, Pella J, Resetar J, Dubikova M, Palinsky M and Bodnar J (1990) [Does the left atrial dimension predict the short-term effect of electric cardioversion?]. Vnitr Lek 36, 753-758
4. Fernandes F, Barroso MF, De Simone A et al (2022) Multi-target neuroprotective effects of herbal medicines for Alzheimer's disease. J Ethnopharmacol 290, 115107
5. Zhao S and Iyengar R (2012) Systems pharmacology: network analysis to identify multiscale mechanisms of drug action. Annu Rev Pharmacol Toxicol 52, 505-521
6. Hopkins AL (2008) Network pharmacology: the next paradigm in drug discovery. Nat Chem Biol 4, 682-690
7. Kibble M, Saarinen N, Tang J, Wennerberg K, Makela S and Aittokallio T (2015) Network pharmacology applications to map the unexplored target space and therapeutic potential of natural products. Nat Prod Rep 32, 1249-1266
8. Huang C, Zheng C, Li Y, Wang Y, Lu A and Yang L (2014) Systems pharmacology in drug discovery and therapeutic insight for herbal medicines. Brief Bioinform 15, 710-733
9. Casas AI, Hassan AA, Larsen SJ et al (2019) From single drug targets to synergistic network pharmacology in ischemic stroke. Proc Natl Acad Sci U S A 116, 7129-7136
10. Wang X, Zhu H, Jiang Y et al (2022) PRODeepSyn: predicting anticancer synergistic drug combinations by embedding cell lines with protein-protein interaction network. Brief Bioinform 23
11. Xu Z (2011) Modernization: One step at a time. Nature 480, S90-92
12. Li P, Zhang H, Zhang W et al (2022) TMNP: a transcriptome-based multi-scale network pharmacology platform for herbal medicine. Brief Bioinform 23
13. Lamb J, Crawford ED, Peck D et al (2006) The Connectivity Map: using gene-expression signatures to connect small molecules, genes, and disease. Science 313, 1929-1935
14. Subramanian A, Narayan R, Corsello SM et al (2017) A Next Generation Connectivity Map: L1000 Platform and the First 1,000,000 Profiles. Cell 171, 1437-1452 e1417
15. Kwon OS, Kim W, Cha HJ and Lee H (2019) In silico drug repositioning: from large-scale transcriptome data to therapeutics. Arch Pharm Res 42, 879-889
16. Jiang H, Hu C and Chen M (2021) The Advantages of Connectivity Map Applied in Traditional Chinese Medicine. Front Pharmacol 12, 474267
17. Lv C, Wu X, Wang X et al (2017) The gene expression profiles in response to 102 traditional Chinese medicine (TCM) components: a general template for research on TCMs. Sci Rep 7, 352
18. Li P, Chen C, Zhang W et al (2019) Detection of Vasodilators From Herbal Components by a

Transcriptome-Based Functional Gene Module Reference Approach. Front Pharmacol 10, 1144
19. Wang L, Zhang W, Lu Z et al (2020) Functional Gene Module-Based Identification of Phillyrin as an Anticardiac Fibrosis Agent. Front Pharmacol 11, 1077
20. Fang S, Dong L, Liu L et al (2021) HERB: a high-throughput experiment- and reference-guided database of traditional Chinese medicine. Nucleic Acids Res 49, D1197-D1206
21. Huang L, Xie D, Yu Y et al (2018) TCMID 2.0: a comprehensive resource for TCM. Nucleic Acids Res 46, D1117-D1120
22. Ru J, Li P, Wang J et al (2014) TCMSP: a database of systems pharmacology for drug discovery from herbal medicines. J Cheminform 6, 13
23. Liu Z, Guo F, Wang Y et al (2016) BATMAN-TCM: a Bioinformatics Analysis Tool for Molecular mechANism of Traditional Chinese Medicine. Sci Rep 6, 21146
24. Wu Y, Zhang F, Yang K et al (2019) SymMap: an integrative database of traditional Chinese medicine enhanced by symptom mapping. Nucleic Acids Res 47, D1110-D1117
25. Kim S, Chen J, Cheng T et al (2021) PubChem in 2021: new data content and improved web interfaces. Nucleic Acids Res 49, D1388-D1395
26. Daina A, Michielin O and Zoete V (2017) SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Sci Rep 7, 42717
27. Daina A, Michielin O and Zoete V (2019) SwissTargetPrediction: updated data and new features for efficient prediction of protein targets of small molecules. Nucleic Acids Res 47, W357-W364
28. Szklarczyk D, Santos A, von Mering C, Jensen LJ, Bork P and Kuhn M (2016) STITCH 5: augmenting protein-chemical interaction networks with tissue and affinity data. Nucleic Acids Res 44, D380-384
29. Chen X, Ji ZL and Chen YZ (2002) TTD: Therapeutic Target Database. Nucleic Acids Res 30, 412-415
30. Yu H, Chen J, Xu X et al (2012) A systematic prediction of multiple drug-target interactions from chemical, genomic, and pharmacological data. PLoS One 7, e37608
31. Zhao S and Li S (2010) Network-based relating pharmacological and genomic spaces for drug target identification. PLoS One 5, e11764
32. UniProt C (2021) UniProt: the universal protein knowledgebase in 2021. Nucleic Acids Res 49, D480-D489
33. Stelzer G, Rosen N, Plaschkes I et al (2016) The GeneCards Suite: From Gene Data Mining to Disease Genome Sequence Analyses. Curr Protoc Bioinformatics 54, 1 30 31-31 30 33
34. Kanehisa M, Furumichi M, Tanabe M, Sato Y and Morishima K (2017) KEGG: new perspectives on genomes, pathways, diseases and drugs. Nucleic Acids Res 45, D353-D361
35. Jassal B, Matthews L, Viteri G et al (2020) The reactome pathway knowledgebase. Nucleic Acids Res 48, D498-D503
36. The Gene Ontology C (2019) The Gene Ontology Resource: 20 years and still GOing strong.

Nucleic Acids Res 47, D330-D338
37. Subramanian A, Tamayo P, Mootha VK et al (2005) Gene set enrichment analysis: a knowledge-based approach for interpreting genome-wide expression profiles. Proc Natl Acad Sci U S A 102, 15545-15550
38. Szklarczyk D, Gable AL, Nastou KC et al (2021) The STRING database in 2021: customizable protein-protein networks, and functional characterization of user-uploaded gene/measurement sets. Nucleic Acids Res 49, D605-D612
39. Keshava Prasad TS, Goel R, Kandasamy K et al (2009) Human Protein Reference Database--2009 update. Nucleic Acids Res 37, D767-772
40. Athanasios A, Charalampos V, Vasileios T and Ashraf GM (2017) Protein-Protein Interaction (PPI) Network: Recent Advances in Drug Discovery. Curr Drug Metab 18, 5-10
41. Pinero J, Ramirez-Anguita JM, Sauch-Pitarch J et al (2020) The DisGeNET knowledge platform for disease genomics: 2019 update. Nucleic Acids Res 48, D845-D855
42. Amberger JS, Bocchini CA, Scott AF and Hamosh A (2019) OMIM.org: leveraging knowledge across phenotype-gene relationships. Nucleic Acids Res 47, D1038-D1043
43. Kohler S, Gargano M, Matentzoglu N et al (2021) The Human Phenotype Ontology in 2021. Nucleic Acids Res 49, D1207-D1217
44. Dancey JE and Chen HX (2006) Strategies for optimizing combinations of molecularly targeted anticancer agents. Nat Rev Drug Discov 5, 649-659
45. Reddy AS and Zhang S (2013) Polypharmacology: drug discovery for the future. Expert Rev Clin Pharmacol 6, 41-47
46. Li S, Zhang B and Zhang N (2011) Network target for screening synergistic drug combinations with application to traditional Chinese medicine. BMC Systems Biology 5, S10
47. Wang X, Xu X, Tao W, Li Y, Wang Y and Yang L (2012) A systems biology approach to uncovering pharmacological synergy in herbal medicines with applications to cardiovascular disease. Evid Based Complement Alternat Med 2012, 519031
48. Li P, Chen J, Zhang W, Fu B and Wang W (2017) Transcriptome inference and systems approaches to polypharmacology and drug discovery in herbal medicine. J Ethnopharmacol 195, 127-136
49. Wang N, Li P, Hu X et al (2019) Herb Target Prediction Based on Representation Learning of Symptom related Heterogeneous Network. Comput Struct Biotechnol J 17, 282-290
50. Zhao W, Wu H and He J (2021) HGNA-HTI: Heterogeneous graph neural network with attention mechanism for prediction of herb-target interactions.
51. Keum J, Yoo S, Lee D and Nam H (2016) Prediction of compound-target interactions of natural products using large-scale drug and protein information. BMC Bioinformatics 17 Suppl 6, 219
52. Grover A and Leskovec J (2016) node2vec: Scalable Feature Learning for Networks. KDD 2016, 855-864
53. Bleakley K and Yamanishi Y (2009) Supervised prediction of drug-target interactions using bipartite local models. Bioinformatics 25, 2397-2403

54. Xu X (2006) New concepts and approaches for drug discovery based on traditional Chinese medicine. Drug Discov Today Technol 3, 247-253
55. Yoo S, Nam H and Lee D (2018) Phenotype-oriented network analysis for discovering pharmacological effects of natural compounds. Sci Rep 8, 11667
56. Yoo S, Yang HC, Lee S et al (2020) A Deep Learning-Based Approach for Identifying the Medicinal Uses of Plant-Derived Natural Compounds. Front Pharmacol 11, 584875
57. Yoo S, Kim K, Nam H and Lee D (2018) Discovering Health Benefits of Phytochemicals with Integrated Analysis of the Molecular Network, Chemical Properties and Ethnopharmacological Evidence. Nutrients 10
58. Kim E, Choi AS and Nam H (2019) Drug repositioning of herbal compounds via a machine-learning approach. BMC Bioinformatics 20, 247
59. Gottlieb A, Stein GY, Ruppin E and Sharan R (2011) PREDICT: a method for inferring novel drug indications with application to personalized medicine. Mol Syst Biol 7, 496
60. Li P, Chen J, Wang J et al (2014) Systems pharmacology strategies for drug discovery and combination with applications to cardiovascular diseases. J Ethnopharmacol 151, 93-107
61. Cases M and Mestres J (2009) A chemogenomic approach to drug discovery: focus on cardiovascular diseases. Drug Discov Today 14, 479-485
62. Kong DX, Li XJ and Zhang HY (2009) Where is the hope for drug discovery? Let history tell the future. Drug Discov Today 14, 115-119
63. Wang Y, Yang H, Chen L, Jafari M and Tang J (2021) Network-based modeling of herb combinations in traditional Chinese medicine. Brief Bioinform 22
64. Wishart DS, Feunang YD, Guo AC et al (2018) DrugBank 5.0: a major update to the DrugBank database for 2018. Nucleic Acids Res 46, D1074-D1082
65. Thorn CF, Klein TE and Altman RB (2005) PharmGKB: the pharmacogenetics and pharmacogenomics knowledge base. Methods Mol Biol 311, 179-191
66. Ye H, Ye L, Kang H et al (2011) HIT: linking herbal active ingredients to targets. Nucleic Acids Res 39, D1055-1059
67. Kuhn M, Letunic I, Jensen LJ and Bork P (2016) The SIDER database of drugs and side effects. Nucleic Acids Res 44, D1075-1079
68. Rappaport N, Twik M, Plaschkes I et al (2017) MalaCards: an amalgamated human disease compendium with diverse clinical and genetic annotation and structured search. Nucleic Acids Res 45, D877-D887
69. Kilicoglu H, Shin D, Fiszman M, Rosemblat G and Rindflesch TC (2012) SemMedDB: a PubMed-scale repository of biomedical semantic predications. Bioinformatics 28, 3158-3160
70. Zhou X, Menche J, Barabasi AL and Sharma A (2014) Human symptoms-disease network. Nat Commun 5, 4212
71. Hu Z, Dong Y, Wang K and Sun Y (2020) Heterogeneous Graph Transformer.
72. Yang K, Liu G, Wang N et al (2018) Heterogeneous network propagation for herb target identification. BMC Med Inform Decis Mak 18, 17
73. Li B, Ma C, Zhao X et al (2018) YaTCM: Yet another Traditional Chinese Medicine Database

for Drug Discovery. Comput Struct Biotechnol J 16, 600-610
74. Xu HY, Liu ZM, Fu Y et al (2017) [Exploiture and application of an internet-based Computation Platform for Integrative Pharmacology of Traditional Chinese Medicine]. Zhongguo Zhong Yao Za Zhi 42, 3633-3638
75. Chen X, Zhou H, Liu YB et al (2006) Database of traditional Chinese medicine and its application to studies of mechanism and to prescription validation. Br J Pharmacol 149, 1092-1103
76. Chen CY (2011) TCM Database@Taiwan: the world's largest traditional Chinese medicine database for drug screening in silico. PLoS One 6, e15939
77. Yoo S, Ha S, Shin M, Noh K, Nam H and Lee D (2018) A Data-Driven Approach for Identifying Medicinal Combinations of Natural Products. IEEE Access 6, 58106-58118
78. Davis AP, King BL, Mockus S et al (2011) The Comparative Toxicogenomics Database: update 2011. Nucleic Acids Res 39, D1067-1072
79. Gunther S, Kuhn M, Dunkel M et al (2008) SuperTarget and Matador: resources for exploring drug-target relationships. Nucleic Acids Res 36, D919-922
80. Chatr-Aryamontri A, Breitkreutz BJ, Oughtred R et al (2015) The BioGRID interaction database: 2015 update. Nucleic Acids Res 43, D470-478
81. Tatonetti NP, Ye PP, Daneshjou R and Altman RB (2012) Data-driven prediction of drug effects and interactions. Sci Transl Med 4, 125ra131
82. UniProt C (2015) UniProt: a hub for protein information. Nucleic Acids Res 43, D204-212
83. Griffith M, Griffith OL, Coffman AC et al (2013) DGIdb: mining the druggable genome. Nat Methods 10, 1209-1210
84. Robinson PN, Kohler S, Bauer S, Seelow D, Horn D and Mundlos S (2008) The Human Phenotype Ontology: a tool for annotating and analyzing human hereditary disease. Am J Hum Genet 83, 610-615
85. Pinero J, Bravo A, Queralt-Rosinach N et al (2017) DisGeNET: a comprehensive platform integrating information on human disease-associated genes and variants. Nucleic Acids Res 45, D833-D839
86. Liu T, Lin Y, Wen X, Jorissen RN and Gilson MK (2007) BindingDB: a web-accessible database of experimentally determined protein-ligand binding affinities. Nucleic Acids Res 35, D198-201
87. Cheng F, Kovacs IA and Barabasi AL (2019) Network-based prediction of drug combinations. Nat Commun 10, 1197
88. Chavan P, Joshi K and Patwardhan B (2006) DNA microarrays in herbal drug research. Evid Based Complement Alternat Med 3, 447-457
89. Hwang YH, Kim T, Cho WK, Jang D, Ha JH and Ma JY (2012) Food- and gender-dependent pharmacokinetics of paeoniflorin after oral administration with Samul-tang in rats. J Ethnopharmacol 142, 161-167
90. Jong Hoon R and Soo C (2001) Y. The effects of Sa-Mul-Tang, a traditional Chinese medicine, on phenylhydrazineinduced anemic rats. The Journal of Applied Phamacology 9, 109-114

91. Wen Z, Wang Z, Wang S et al (2011) Discovery of molecular mechanisms of traditional Chinese medicinal formula Si-Wu-Tang using gene expression microarray and connectivity map. PLoS One 6, e18278
92. Liu M, Fan J, Wang S et al (2013) Transcriptional profiling of Chinese medicinal formula Si-Wu-Tang on breast cancer cells reveals phytoestrogenic activity. BMC Complement Altern Med 13, 11
93. Fang Z, Lu B, Liu M et al (2013) Evaluating the pharmacological mechanism of Chinese medicine Si-Wu-Tang through multi-level data integration. PLoS One 8, e72334
94. Huang GC, Tsai YZ, Lee CJ, Chang HY and Wang CC (2019) Elucidation of the Effects of Si-Wu Tang on Menstrual Disorder Patterns through Activation of Aromatase and Antioxidation. Evid Based Complement Alternat Med 2019, 4761651
95. Kim J and You S (2020) Samul-tang ameliorates oocyte damage due to cyclophosphamide-induced chronic ovarian dysfunction in mice. Sci Rep 10, 21925
96. Kim J and You S (2021) Effect of samul-tang on female fertility via RAS signaling pathway in ovaries of aged mice. Aging (Albany NY) 13, 14829-14842
97. Kim J and You S (2022) Restoration of miR-223-3p expression in aged mouse uteri with Samul-tang administration. Integr Med Res 11, 100835
98. Kim J, Fann DY, Seet RC, Jo DG, Mattson MP and Arumugam TV (2016) Phytochemicals in Ischemic Stroke. Neuromolecular Med 18, 283-305
99. Duan X, Han L, Peng D et al (2018) High Throughput mRNA Sequencing Reveals Potential Therapeutic Targets of Tao-Hong-Si-Wu Decoction in Experimental Middle Cerebral Artery Occlusion. Front Pharmacol 9, 1570
100. Qu Z, Cui J, Harata-Lee Y et al (2016) Identification of candidate anti-cancer molecular mechanisms of Compound Kushen Injection using functional genomics. Oncotarget 7, 66003-66019
101. Shen H, Qu Z, Harata-Lee Y et al (2019) Understanding the Mechanistic Contribution of Herbal Extracts in Compound Kushen Injection With Transcriptome Analysis. Front Oncol 9, 632
102. Nourmohammadi S, Aung TN, Cui J et al (2019) Effect of Compound Kushen Injection, a Natural Compound Mixture, and Its Identified Chemical Components on Migration and Invasion of Colon, Brain, and Breast Cancer Cell Lines. Front Oncol 9, 314
103. Cui J, Qu Z, Harata-Lee Y et al (2020) The effect of compound kushen injection on cancer cells: Integrated identification of candidate molecular mechanisms. PLoS One 15, e0236395
104. Zhang L, Ji YX, Jiang WL and Lv CJ (2015) Protective roles of pulmonary rehabilitation mixture in experimental pulmonary fibrosis in vitro and in vivo. Braz J Med Biol Res 48, 545-552
105. Li H, Wang Z, Zhang J et al (2018) Feifukang ameliorates pulmonary fibrosis by inhibiting JAK-STAT signaling pathway. BMC Complement Altern Med 18, 234
106. Tan YQ, Chen HW, Li J and Wu QJ (2020) Efficacy, Chemical Constituents, and Pharmacological Actions of Radix Paeoniae Rubra and Radix Paeoniae Alba. Front Pharmacol 11, 1054

107. Baek SJ, Lee H, Park SM et al (2022) Identification of a novel anticancer mechanism of Paeoniae Radix extracts based on systematic transcriptome analysis. Biomed Pharmacother 148, 112748
108. Chen B, Garmire L, Calvisi DF, Chua MS, Kelley RK and Chen X (2020) Harnessing big 'omics' data and AI for drug discovery in hepatocellular carcinoma. Nat Rev Gastroenterol Hepatol 17, 238-251
109. Luo S, Li H, Mo Z et al (2019) Connectivity map identifies luteolin as a treatment option of ischemic stroke by inhibiting MMP9 and activation of the PI3K/Akt signaling pathway. Exp Mol Med 51, 1-11
110. Liu J, Lee J, Salazar Hernandez MA, Mazitschek R and Ozcan U (2015) Treatment of obesity with celastrol. Cell 161, 999-1011
111. Wang TY, Li Q and Bi KS (2018) Bioactive flavonoids in medicinal plants: Structure, activity and biological fate. Asian J Pharm Sci 13, 12-23
112. Zhu Q, Wong AK, Krishnan A et al (2015) Targeted exploration and analysis of large cross-platform human transcriptomic compendia. Nat Methods 12, 211-214, 213 p following 214
113. Lee JJ, Kim KH, Kim EJ et al (2018) Anti-inflammatory activity of the decoction of Forsythia suspensa (Thunb.) Vahl is related to Nrf2 and A20. J Ethnopharmacol 227, 97-104
114. Belyaeva A, Cammarata L, Radhakrishnan A et al (2021) Causal network models of SARS-CoV-2 expression and aging to identify candidates for drug repurposing. Nat Commun 12, 1024
115. Budhraja A, Basu A, Gheware A et al (2022) Molecular signature of postmortem lung tissue from COVID-19 patients suggests distinct trajectories driving mortality. Dis Model Mech 15