Original Article

Network Pharmacology Analysis Uncovers the Potential Anti-Hypertensive Mechanisms of Xia Sang Ju Granule

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Abstract

Background and objectives: Xia Sang Ju (XSJ) granule, a Chinese drug and herbal tea made up of *Prunellae spica* (Xia Ku Cao), *Mori folium* (Sang Ye), and *Flos Chrysanthemi Indici* (Ye Ju Hua), is commonly used for fever, headache, and sore throat. The underlying pharmacological mechanism of XSJ on hypertension treatment is described here, based on network pharmacology.

Methods: The compounds in XSJ were searched using the Traditional Chinese Medicine Systems Pharmacology Database and Analysis Platform (commonly known as TCMSP), and the active components, according to oral bioavailability and drug likeness, were screened. Compounds targets were predicted by the SwissTargetPrediction web server, while hypertension targets were collected from the Online Mendelian Inheritance in Man (commonly known as OMIM) and GeneCards databases. The interaction of targets was analyzed by STRING. The compound-compound target network was constructed by Cytoscape. Gene Ontology enrichment and Kyoto Encyclopedia of Genes and Genomes (commonly known as KEGG) pathways were analyzed by the Database for Annotation, Visualization and Integrated Discovery (commonly known as DAVID).

Results: Forty-five active compounds were obtained from 359 ingredients present in the XSJ decoction, corresponding to 237 targets. In addition, 189 genes were found to be related to hypertension, of which 11 overlapped with XSJ targeted by 28 compounds and were thus considered therapeutically-relevant. *ESR2* was the most frequent gene targeted by the compounds, while *NR3C1* showed the most interaction with other genes. These results revealed that the anti-hypertensive activity of XSJ may directly relate to the regulation of several hypertension-associated biological processes and pathways, such as cellular nitrogen compound biosynthetic process, positive regulation of the nitrogen compound metabolic process, steroid hormone biosynthesis, and aldosterone-regulated sodium reabsorption.

Conclusions: These findings provide a reference for further interpretation of the potential mode of action of XSJ against hypertension and serve as an example for elucidation of the Traditional Chinese Medicine concept of "multiple compounds-multiple targets-multiple effects".

Keywords: Xia Sang Ju granule; Hypertension; Network pharmacology; Multiple compounds-multiple targets-multiple effects.

Abbreviations: DAVID, Database for Annotation, Visualization and Integrated Discovery; KEGG, Kyoto Encyclopedia of Genes and Genomes; OB, oral bioavailability; DL, drug likeness; OMIM, Online Mendelian Inheritance in Man; SMILES, simplified molecular-input line-entry system; TCMSP, Traditional Chinese Medicine Systems Pharmacology Database and Analysis Platform; TCM, traditional Chinese medicine; XSJ, Xia Sang Ju.

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Introduction

Xia Sang Ju (XSJ) granule is a traditional Chinese drug as well as a kind of Chinese herbal tea which is made up of *Prunellae spica* (*Xia Ku Cao*), *Mori folium* (*Sang Ye*), and *Flos Chrysanthemi Indici* (*Ye Ju Hua*). It originates from the classic prescription called *Sang Ju Yin* that was recorded in the *Treatise on Differentiation and Treatment of Epidemic Febrile Diseases (Wen Bing Tiao Bian)* by Wu Jutong in the Qing Dynasty. Though XSJ is well-known for treating fever, headache, and sore throat, hypertension is also one of the main functions of XSJ.[1] However, how XSJ plays a part in anti-hypertensive activity remains

unclear, due to the complexity of Traditional Chinese medicine (TCM).

Hypertension is characterized by elevated blood pressure in arteries, and is the most common of the chronic diseases and one of the most important risk factors for cerebrovascular diseases; causing an estimated 7.5 million deaths, it accounts for 12.8% of the total deaths.[2] So far, commonly-used anti-hypertensive drugs include diuretics, beta-blockers, angiotensin converting enzyme inhibitors, calcium channel blockers, angiotensin receptor blockers, *etc*.[3] Unfortunately, no specific medicine can yet cure high blood pressure.

TCMs are extensively used in eastern countries, as treatments for such chronic diseases as hypertension, diabetes and stroke, and their advantages have been gradually recognized through the increasing number of people who seek natural herbal remedies in western countries.[4] Most existing research is limited to a certain gene target while interpreting the mechanism of a drug, an approach which may ignore the multi-component, multi-target, multi-pathway characteristics of Chinese herbal formulae.[4] Network pharmacology, based on an integrated multidisciplinary concept, is a powerful tool that analyzes the multi-level network of molecular-target-pathway-disease through the interaction between TCM and disease from a holistic perspective.[5–9]

In this study, firstly the active compounds of XSJ were screened computationally, according to oral bioavailability (OB) and drug likeness (DL)[10] and then the potential compound targets and hypertension-related targets were predicted. Finally the XSJ-compound-hypertension networks were constructed, so as to deeply understand the potential underlying mechanism of the anti-hypertensive effect of XSJ.

Materials and methods

Compounds of XSJ

The Traditional Chinese Medicine Systems Pharmacology Database and Analysis Platform (commonly known as TCMSP; http://www.tcmspw.com/tcmsp.php, version 2.3)[11] was used to collect the compound information of XSJ. A total of 60 compounds in *Prunellae spica*, 269 compounds in *Mori folium*, and 30 compounds in *Flos Chrysanthemi Indici* were found. To select the potential active compounds, OB and DL,[10] the most important criteria for drug screening, was set to be ≥30% and ≥0.18, respectively.[12]

Compound targets

To predict the most relevant targets of compounds, the simplified molecular-input line-entry system (referred to as SMILES) format of each compound was input into the SwissTargetPrediction website (http://www.swisstargetprediction.ch/) with the organism limited to *Home sapiens*.[13,14]

Hypertension targets

Genes associated with hypertension were searched from the Online Mendelian Inheritance in Man (commonly known as OMIM) database (http://www.omim.org/)[15,16] and GeneCards database (https://www.genecards.org/)[17] using the keywords "hypertension" or "high blood pressure".

Protein-protein interaction

The STRING database (https://string-db.org/, version 10.5) was used to analysis the protein-protein interaction.[18] Protein names were input and organism was limited to *Homo sapiens*. Data of protein-protein interactions were obtained and saved as TSV files.

GeneMANIA analysis

A weighted composite functional interaction network for hypertension-related genes were constructed by GeneMANIA (https://genemania.org/).[19] Genes of interest were input and organism was limited to *Homo sapiens*.

Network construction

All the networks were constructed by Cytoscape software (https://cytoscape.org/, version 3.6.1).[20]

Gene ontology enrichment analysis

Gene ontology enrichment analysis for biological processes and Kyoto Encyclopedia of Genes and Genomes (commonly known as KEGG) pathways were performed by Database for Annotation, Visualization and Integrated Discovery, commonly known as DAVID, 6.8 server (https://david.ncifcrf.gov/).[21,22]

Results

Screen of active compounds

In total, 359 compounds in XSJ were obtained from the TCMSP database. After filtering by OB and drug likeness parameters, 11 compounds from *Prunellae spica*, 29 compounds from *Mori folium*, and 12 compounds from *Flos Chrysanthemi Indici* with favorable pharmacokinetic profiles were included for further investigation (Table 1). Specifically, beta-sitosterol and quercetin were found in all three of the herbs, and kaempferol as well as stigmasterol were originated from both *Prunellae spica* and *Mori folium,* while luteolin was found in *Prunellae spica* and *Flos Chrysanthemi Indici.*

Hypertension network analysis

In total, 189 genes associated with hypertension were obtained from the OMIM and GeneCards databases after elimination of false positives and repetitive genes (Table s1). The interaction of hypertension target genes was analyzed by GeneMANIA (Fig. 1, Table s2) and a network containing 274 nodes and 10,742 edges was constructed. This result showed that 55.08% of genes were co-expressed, and 20.87% were expressed in the same tissue or their products in the same cellular location. Among the genes, 11.02% were found to be involved in physical interaction, while 4.86% were engaged in predicted functional relationships. Up to 3.61% were identified as possibly participating in the same pathway, 3.01% had shared protein domains, and 1.55% had genetic interactions that were functionally associated.

Table 1. Active compounds in the herbs and their properties

Mol ID	Compound	OB, %	DL	Herbs
MOL000358	Beta-sitosterol	36.91	0.75	*Prunellae spica, Mori folium, Flos Chrysanthemi Indici*
MOL000422	Kaempferol	41.88	0.24	*Prunellae spica, Mori folium*
MOL004355	Spinasterol	42.98	0.76	*Prunellae spica*
MOL000449	Stigmasterol	43.83	0.76	*Prunellae spica, Mori folium*
MOL004798	Delphinidin	40.63	0.28	*Prunellae spica*
MOL000006	Luteolin	36.16	0.25	*Prunellae spica, Flos Chrysanthemi Indici*
MOL006767	Vulgaxanthin-I	56.14	0.26	*Prunellae spica*
MOL006772	Poriferasterol monoglucoside_qt	43.83	0.76	*Prunellae spica*
MOL006774	Stigmast-7-enol	37.42	0.75	*Prunellae spica*
MOL000737	Morin	46.23	0.27	*Prunellae spica*
MOL000098	Quercetin	46.43	0.28	*Prunellae spica, Mori folium, Flos Chrysanthemi Indici*
MOL001771	Poriferast-5-en-3beta-ol	36.91	0.75	*Mori folium*
MOL002218	Scopolin	56.45	0.39	*Mori folium*
MOL002773	Beta-carotene	37.18	0.58	*Mori folium*
MOL003842	Albanol	83.16	0.24	*Mori folium*
MOL003847	Inophyllum E	38.81	0.85	*Mori folium*
MOL003850	26-Hydroxy-dammara-20,24-dien-3-one	44.41	0.79	*Mori folium*
MOL003851	Isoramanone	39.97	0.51	*Mori folium*
MOL003856	Moracin B	55.85	0.23	*Mori folium*
MOL003857	Moracin C	82.13	0.29	*Mori folium*
MOL003858	Moracin D	60.93	0.38	*Mori folium*
MOL003859	Moracin E	56.08	0.38	*Mori folium*
MOL003860	Moracin F	53.81	0.23	*Mori folium*
MOL003861	Moracin G	75.78	0.42	*Mori folium*
MOL003862	Moracin H	74.35	0.51	*Mori folium*
MOL003879	4-Prenylresveratrol	40.54	0.21	*Mori folium*
MOL000433		68.96	0.71	*Mori folium*
MOL000729	Oxysanguinarine	46.97	0.87	*Mori folium*
MOL001439	Arachidonic acid	45.57	0.2	*Mori folium*
MOL001506	Supraene	33.55	0.42	*Mori folium*
MOL003759	Iristectorigenin A	63.36	0.34	*Mori folium*
MOL003975	Icosa-11,14,17-trienoic acid methyl ester	44.81	0.23	*Mori folium*
MOL006630	Norartocarpetin	54.93	0.24	*Mori folium*
MOL007179	Linolenic acid ethyl ester	46.1	0.2	*Mori folium*
MOL007879	Tetramethoxyluteolin	43.68	0.37	*Mori folium*
MOL013083	Skimmin (8CI)	38.35	0.32	*Mori folium*
MOL001689	Acacetin	34.97	0.24	*Flos Chrysanthemi Indici*
MOL001790	Linarin	39.84	0.71	*Flos Chrysanthemi Indici*
MOL000359	Sitosterol	36.91	0.75	*Flos Chrysanthemi Indici*
MOL008173	Daucosterol_qt	36.91	0.75	*Flos Chrysanthemi Indici*

Table 1. Active compounds in the herbs and their properties - *(continued)*

Mol ID	Compound	OB, %	DL	Herbs
MOL008915	Acacetin-7-O-β-D-galactopyranoside	50.19	0.77	*Flos Chrysanthemi Indici*
MOL008918	Arteglasin A	52.45	0.33	*Flos Chrysanthemi Indici*
MOL008919	(2S,6S,7aR)-2-[(1E,3E,5E,7E,9E,11E,13E,15E)-16-[(4S)-4-Hydroxy-2,6,6-trimethyl-1-cyclohexenyl]-1,5,10,14-tetramethylhexadeca-1,3,5,7,9,11,13,15-octaenyl]-4,4,7a-trimethyl-2,5,6,7-tetrahydrobenzofuran-6-ol	59.52	0.55	*Flos Chrysanthemi Indici*
MOL008924	Azuleno(4,5-b)furan-2(3H)-one, 4-(acetyloxy)-3a,4,5,6,6a,7,9a,9b-octahydro-6-hydroxy-6,9-dimethyl-3-methylene-, (3aR-(3aalpha, 4alpha,6alpha,6aalpha,9aalpha,9bbeta))-	68.44	0.27	*Flos Chrysanthemi Indici*
MOL008925	(3aR,4S,6R,6aR,9aR,9bR)-4,6-Dihydroxy-6,9-dimethyl-3-methylene-4,5,6a,7,9a,9b-hexahydro-3aH-azuleno[5,4-d]furan-2-one	40.08	0.19	*Flos Chrysanthemi Indici*

DL, drug likeness.

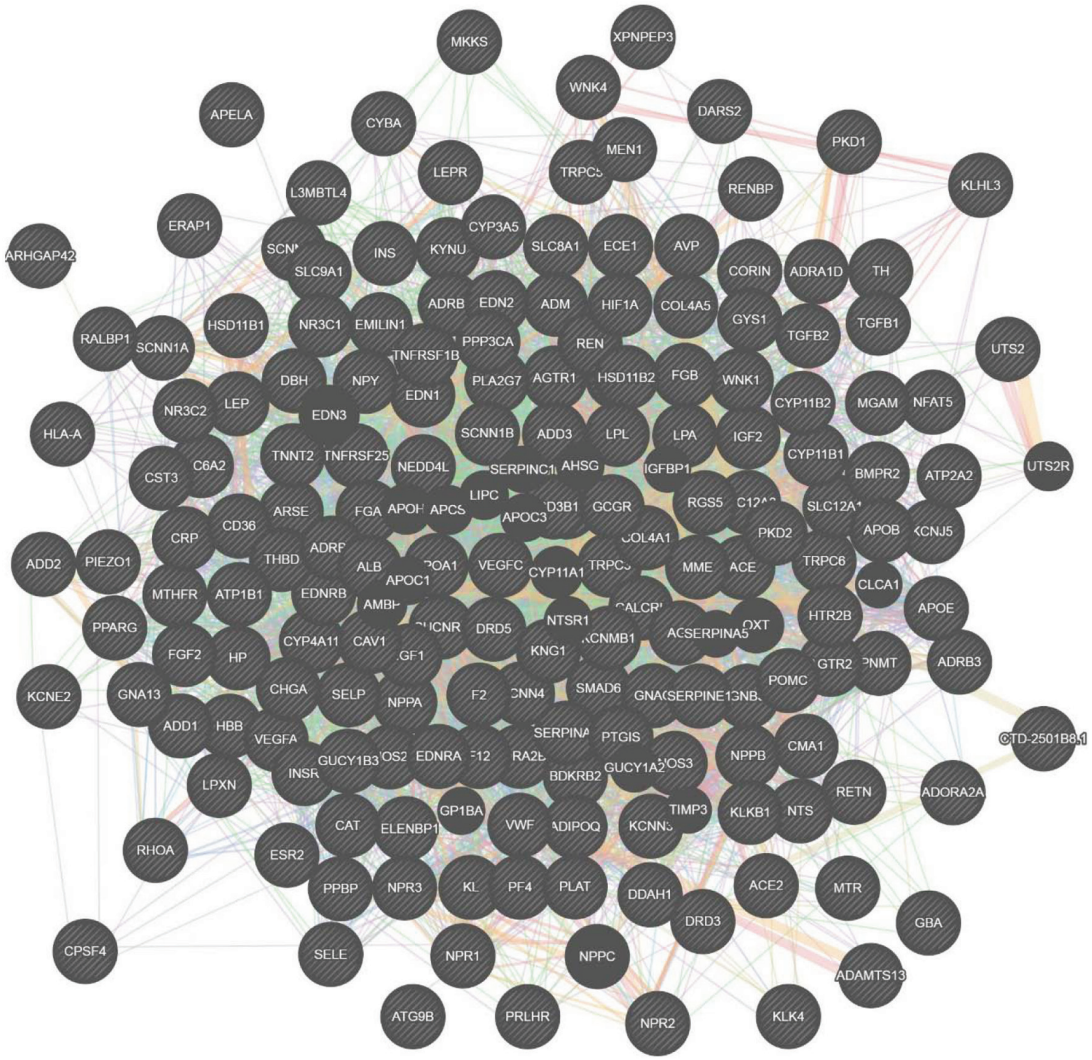

Fig. 1. Protein-protein interaction network of hypertension targets.

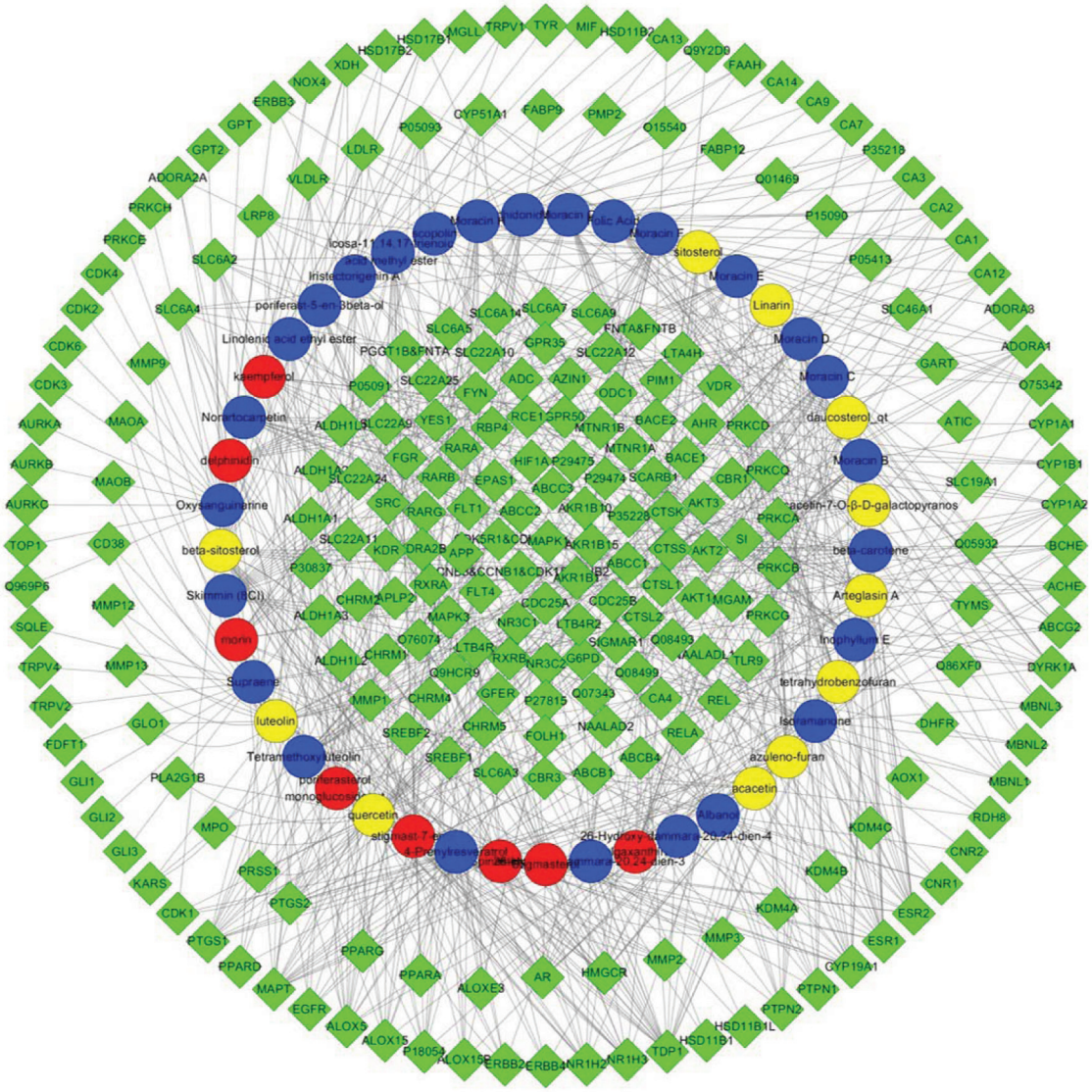

Fig. 2. Compound-compound target network of Xia Sang Ju (XSJ). Compound targets are denoted by green hexagons; *Prunellae spica* by blue circles; *Mori folium* by red circles; *Flos Chrysanthemi Indici* by yellow circles. "Azuleno(4,5-b)furan-2(3H)-one, 4-(acetyloxy)-3a,4,5,6,6a,7,9a,9b-octahydro-6-hydroxy-6,9-dimethyl-3-methylene-, (3aR-(3aalpha,4alpha,6alpha,6aalpha,9aalpha,9bbeta))" is abbreviated to "Azuleno-furan"; "(2S,6S,7aR)-2-[(1E,3E,5E,7E,9E,11E,13E,15E)-16-[(4S)-4-hydroxy-2,6,6-trimethyl-1-cyclohexenyl]-1,5,10,14-tetramethylhexadeca-1,3,5,7,9,11,13,15-octaenyl]-4,4,7a-trimethyl-2,5,6,7-tetrahydrobenzofuran-6-ol)" is abbreviated to "tetrahydrobenzofuran"; "(3aR,4S,6R,6aR,9aR,9bR)-4,6-dihydroxy-6,9-dimethyl-3-methylene-4,5,6a,7,9a,9b-hexahydro-3aH-azuleno[5,4-d]furan-2-one" is abbreviated to "azuleno-furan".)

Analysis of compound-compound target network

The SMILES format of each compound was input into SwissTargetPrediction, and predicted compound targets were obtained (Table s3). A compound-compound target network was constructed, consisting of 282 nodes and 703 edges (Fig. 2). These results showed that some target genes may be modulated by many compounds, such as the *ESR1*, *AR*, *MAPT*, *CYP19A1*, and *HMGCR* genes. While the *AOX1*, *CTSK*, *OCD1*, *SRC*, *RARA*, *NOX4*, and *CDC25B* genes are hit by only one compound. Interestingly, both SLC6A4 and P05093 can be regulated by poriferast-5-en-3beta-ol, beta-sitosterol, poriferasterol mo noglucoside_qt, stigmast-7-enol, spinasterol, daucosterol_qt, and sitosterol. Both the *NR1H2* and *NR1H3* genes can be modulated by 26-hydroxy-dammara-20,24-dien-3, poriferast-5-en-3beta-ol, beta-sitosterol, poriferasterol monoglucoside_qt, stigmast-7-enol, spinasterol, stigmasterol, daucosterol_qt, and sitosterol. This predicted compound-compound target network strengthens the concepts of multi-compound-multi-target of TCM, in which different active components in XSJ may regulate the same targets and one active ingredient may also modulate various targets.

Hypertension-related compound target network analysis

Eleven genes with commonalities between hypertension genes and compound targets were found and a hypertension-related compound

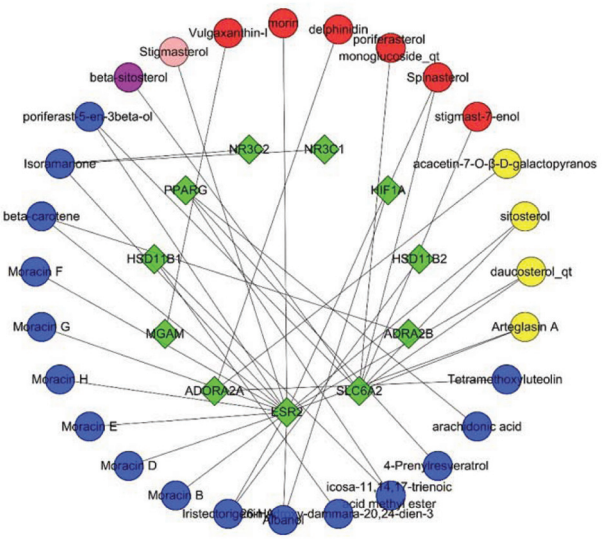

Fig. 3. XSJ-hypertension network. Compound targets and hypertension targets are denoted by green hexagons; *Prunellae spica* by blue circles; *Mori folium* by red circles; *Flos Chrysanthemi Indici* by yellow circles; *Prunellae spica* and *Mori folium* by pink circle; *Prunellae spica* and *Mori folium* and *Flos Chrysanthemi Indici* by purple circle.

target network was constructed (Fig. 3, Table 2), which contained 39 nodes and 39 edges. Among the 28 compounds directly interacting with these genes, 8 of them came from *Prunellae spica*, 18 were from *Mori folium*, and 5 were from *Flos Chrysanthemi Indici*. The protein classes for the 11 common genes were obtained from the DisGeNET database. The XSJ and hypertension-related targets' protein-protein interaction network is shown in Figure 4. *ESR2* and *SLC6A2*, both of which play a role in nucleic acid binding, as receptor and transcription factor, or transporter, were the most frequent genes targeted by the compounds. ESR2 and SLC6A2 are known to be important to cardiovascular physiology and blood pressure regulation.[23–27] These results suggested that the anti-hypertension effect of XSJ may be regulated mainly by ESR2 and SLC6A2 (Table 3).

Biological functional analysis

Biological functions of the hypertension-related compound targets were annotated to explain the possible mode of action of XSJ in hypertension. Gene ontology enrichment analysis was performed on the 11targets by DAVID. The top five biological processes were cellular nitrogen compound biosynthetic process, organic cyclic compound biosynthetic process, aromatic compound biosynthetic process, heterocycle biosynthetic process, and nucleobase-containing compound biosynthetic process (Fig. 5a). The significant KEGG pathways included neuroactive ligand-receptor interaction, steroid hormone biosynthesis, aldosterone-regulated sodium reabsorption, PPAR signaling pathway, and thyroid cancer (Fig. 5b). These results elucidated that XSJ may exert anti-hypertension activity through multi-biological processes as well as multi-pathways.

Discussion

The escalation of hypertension cases global effects. Coupled with lack of any promising hypotensor, this then requires multiple approaches for treatment, including lifestyle modifications and new drugs. Though XSJ is generally used for treatment of fever, headache, sore throat, and as a beverage for clearing heat, hypertension is also one of the major functions.[1] Nevertheless, the mechanism of action for XSJ working on hypertension remains to be fully understood.

During the development of hypertension, endothelin, nitric oxide, and angiotensin II are key factors. Vascular endothelial cells can produce both systolic and vasoactive substances for maintaining vasomotor balance and normal tension. Endothelin is the strongest vasoconstrictor and promotes smooth muscle proliferation,[28] while nitric oxide is the main vasodilator substance released by vascular endothelial cells. Endothelin harbors angiotensin-converting enzyme activity that catalyzes the synthesis of angiotensin II; however, angiotensin II can induce expression of the endothelin gene in endothelial cells. Nitric oxide inhibits the production and release of endothelin, and also inhibits the release of renin, which in turn inhibits the production of angiotensin II.[29–31]

Despite few publications in the publicly available literature describing the anti-hypertension activity of XSJ so far, recent studies have proven that the extracts and some compounds of all three herbs in XSJ have direct or indirect anti-hypertensive effect, consistent with some of the biological processes found in our study. Ethanol extract of *Prunella vulgaris L* has been shown to increase the content of nitric oxide, to decrease the content of endothelin and angiotensin II, and finally to reduce blood pressure significantly in a spontaneously hypertensive rat model (*e.g.,* positive regulation of the nitric oxide biosynthetic process, regulation of the systemic arterial blood pressure by endothelin).[32] Flavonoid compounds *in Mori folium* have also been found to expand the coronary vessels, improve myocardial circulation, and reduce blood pressure (*e.g.,* regulation of blood pressure).[33] Ethanol extract of *Flos Chrysanthemi Indici* has shown hypotensive effect in clinical studies.[34] Intriguingly, luteolin from *Prunella vulgaris L and Flos Chrysanthemi Indici* might inhibit vascular smooth muscle cell proliferation and migration, which is pivotal in the development of arterial remodeling during hypertension (*e.g.,* blood vessel remodeling), by suppressing transforming growth factor-β receptor 1 signaling.[35]

Luteolin can ameliorate hypertensive vascular remodeling through inhibition of proliferation and migration of angiotensin II-induced vascular smooth muscle cells, a process that is mediated by the regulation of MAPK signaling pathway and the production of reactive oxygen species (*e.g.,* blood vessel remodeling, positive regulation of the reactive oxygen species metabolic process).[36] Quercetin from *Prunella vulgaris L and Flos Chrysanthemi Indici* can attenuate hypertension via reduction in oxidative stress and improving endothelial function, as shown in an acute fluoride-induced hypertension and cardiovascular complications model.[37] Furthermore, quercetin was shown to reduce hypertension-induced vascular remodeling, oxidative stress and MMP-2 activity in aortas in the two-kidney one-clip hypertensive Wistar rat model (*e.g.,* blood vessel remodeling).[38] Quercetin can also attenuate vascular contraction through the LKB1-AMPK signaling pathway (*e.g.,* regulation of vasoconstriction).[39] Delphinidin and quercetin were shown to block the renin-angiotensin system signaling pathway through inhibition of angiotensin-converting enzyme activity and decreasing the production of its mRNA.[40] Finally, linarin from *Flos Chrysanthemi Indici* was shown to directly or indirectly activate macrophages and affect the inhibition of nitric oxide that is responsible for vasodilation and hypotension (*e.g.,* vasodilation).[41]

The current study provided a prediction of the potential mechanism of XSJ as treatment of hypertension, based on a computational approach. There are some limitations of this work. First, the

Table 2. Candidate compounds from Xia Sang Ju and their potential targets associated with hypertension

No.	Compound	Target gene code	Herbs
1	Stigmast-7-enol	*SLC6A2*	*Prunellae spica*
2	Spinasterol	*SLC6A2, ESR2*	*Prunellae spica*
3	Poriferasterol monoglucoside_qt	*SLC6A2*	*Prunellae spica*
4	Delphinidin	*ADORA2A*	*Prunellae spica*
5	Morin	*ESR2*	*Prunellae spica*
6	Stigmasterol	*ESR2*	*Prunellae spica, Mori folium*
7	Vulgaxanthin-I	*MGAM*	*Prunellae spica*
8	Beta-sitosterol	*SLC6A2*	*Prunellae spica, Mori folium, Flos Chrysanthemi Indici*
9	Poriferast-5-en-3beta-ol	*SLC6A2, ESR2*	*Mori folium*
10	Isoramanone	*ESR2, NR3C2, NR3C1*	*Mori folium*
11	Beta-carotene	*ADRA2B, ESR2*	*Mori folium*
12	Moracin F	*ESR2*	*Mori folium*
13	Moracin G	*ESR2*	*Mori folium*
14	Moracin H	*ESR2*	*Mori folium*
15	Moracin E	*ESR2*	*Mori folium*
16	Moracin D	*ESR2*	*Mori folium*
17	Moracin B	*ESR2*	*Mori folium*
18	Iristectorigenin A	*ESR2, HSD11B2*	*Mori folium*
19	Albanol	*HIF1A, ESR2*	*Mori folium*
20	26-Hydroxy-dammara-20,24-dien-3	*HSD11B1*	*Mori folium*
21	Icosa-11,14,17-trienoic acid methyl ester	*HSD11B1, PPARG*	*Mori folium*
22	4-Prenylresveratrol	*PPARG*	*Mori folium*
23	Arachidonic acid	*PPARG*	*Mori folium*
24	Tetramethoxyluteolin	*ADORA2A*	*Mori folium*
25	Arteglasin A	*SLC6A2, ESR2*	*Flos Chrysanthemi Indici*
26	Daucosterol_qt	*SLC6A2, ESR2*	*Flos Chrysanthemi Indici*
27	Sitosterol	*SLC6A2, ESR2*	*Flos Chrysanthemi Indici*
28	Acacetin-7-O-β-D-galactopyranos	*ADORA2A*	*Flos Chrysanthemi Indici*

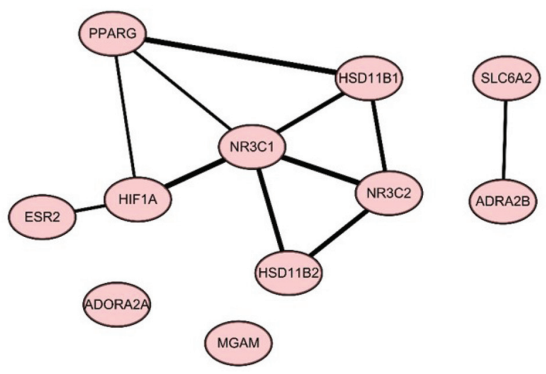

Fig. 4. XSJ and hypertension-related targets' protein-protein interaction network.

components in TCM herbs have not yet been completely identified; thus, the databases of compounds are not complete, precluding their ability to represent the integral spectrum of compounds responsible for the anti-hypertension effect. Second, all of the data were based on *silico* analysis, and as such there may be many false positive and false negative interactions between the found compound-protein and protein-protein interactions. What's more, the relationship between XSJ and anti-hypertension activity was identified by enrichment analysis. Therefore, the associations presented herein should be further investigated for experimental verification to achieve more accurate and reliable inferences in the future.

Future directions

The associated biological processes and pathways need further

Table 3. Hypertension-related targets of Xia Sang Ju

No.	Target	Uniprot ID	Gene code	Protein class	Frequency
1	Estrogen receptor-beta	Q92731	*ESR2*	Nucleic acid binding; receptor; transcription factor	17
2	Solute carrier family 6 member 2	P23975	*SLC6A2*	Transporter	8
3	Nuclear receptor subfamily 3, group C, member 1	P04150	*NR3C1*	Nucleic acid binding; receptor; transcription factor	1
4	Nuclear receptor subfamily 3, group C, member 2	P08235	*NR3C2*	Nucleic acid binding; receptor; transcription factor	1
5	Adenosine receptor A2a	P29274	*ADORA2A*	Receptor	3
6	Adrenoceptor alpha 2B	P18089	*ADRA2B*	Receptor	1
7	Maltase-glucoamylase	O43451	*MGAM*	Hydrolase	1
8	Hypoxia-inducible factor 1, alpha subunit	Q16665	*HIF1A*	Nucleic acid binding; transcription factor	1
9	11-Beta-hydroxysteroid dehydrogenase, type I	P28845	*HSD11B1*	None	1
10	Peroxisome proliferator-activated receptor-gamma	P37231	*PPARG*	Nucleic acid binding; receptor; transcription factor	3
11	Corticosteroid 11-beta-dehydrogenase isozyme 2	P80365	*HSD11B2*	Oxidoreductase	1

investigation for confirmation of the exact mechanism of XSJ in hypertension treatment.

Conclusions

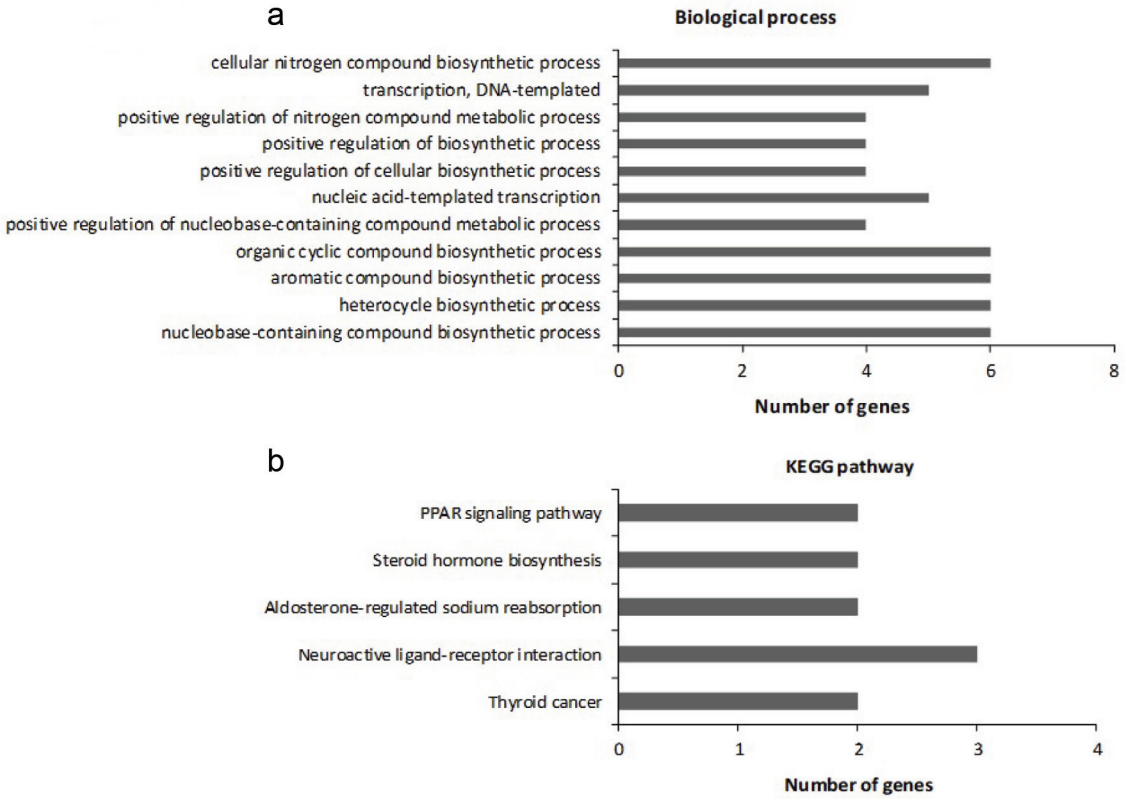

Fig. 5. Gene ontology functional analysis. (a) Biological processes terms. (b) Significant KEGG pathways.

Collectively, the findings presented herein suggest that the compounds in XSJ exert their anti-hypertensive effect via multiple biological processes, such as regulation of blood pressure, blood vessel remodeling, regulation of the nitric oxide biosynthetic process and so on, which is in accord with the TCM therapy concept of "multiple compounds-multiple targets-multiple effects". Though further experiments are needed to verify this finding, this study revealed the potential anti-hypertensive mechanism of XSJ from holistic and systematic perspectives by using network pharmacology.

Supporting information

Supplementary material for this article is available at https://doi.org/10.14218/JERP.2020.00008.

Table s1. Hypertension targets.

Table s2. Interaction of genes related to hypertension.

Table s3. Relationships between compounds and targets.

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Conflict of interest

None.

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