



Network Patterns of Herbal Combinations in Traditional Chinese Clinical Prescriptions

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As a well-established multidrug combinations schema, traditional Chinese medicine (herbal prescription) has been used for thousands of years in real-world clinical settings. This paper uses a complex network approach to investigate the regularities underlying multidrug combinations in herbal prescriptions. Using five collected large-scale real-world clinical herbal prescription datasets, we construct five weighted herbal combination networks with herb as nodes and herbal combinational use in herbal prescription as links. We found that the weight distribution of herbal combinations displays a clear power law, which means that most herb pairs were used in low frequency and some herb pairs were used in very high frequency. Furthermore, we found that it displays a clear linear negative correlation between the clustering coefficients and the degree of nodes in the herbal combination network (HCNet). This indicates that hierarchical properties exist in the HCNet. Finally, we investigate the molecular network interaction patterns between herb related target modules (i.e., subnetworks) in herbal prescriptions using a network-based approach and further explore the correlation between the distribution of herb combinations and prescriptions. We found that the more the hierarchical prescription, the better the corresponding effect. The results also reflected a well-recognized principle called “*Jun-Chen-Zuo-Shi*” in TCM formula theories. This also gives references for multidrug combination development in the field of network pharmacology and provides the guideline for the clinical use of combination therapy for chronic diseases.

Keywords: network pharmacology, complex network, herb combination network, clinical prescription, network pattern

INTRODUCTION

Human disease phenomenon is complicated due to its roots from the disturbance of diverse pathogens on the hierarchical organization of the human life system interacting with the complex natural and social environment. Therefore, finding effective clinical treatments for chronic complex diseases such as diabetes, heart disease, stroke, and cancer is extremely difficult. Furthermore, due to the individualized phenotypes and genotypes incorporated in distinct persons, adverse drug reactions (ADRs) or drug side effects have increasingly become a major health issue when using targeted drugs for patients in real-world clinical settings (Duke et al., 2011; Smyth et al., 2012; Aagaard and Hansen, 2013). For example, there are 106,000 deaths and 2.2 million serious events caused by ADRs in the United States each year (Lazarou et al., 1998), which were once considered as a leading cause of death in the United States (Lazarou et al., 1998). For elderly patients, the ADR is even serious due to the need for multiple prescribed drugs for chronic disease comorbidities (Routledge et al., 2004). However, this problem cannot be well addressed since, for modern drug discovery, the safety of new agents only can be known with certainty if a drug has been on the market for many years. Drugs were only used for selected populations and in limited periods (Lasser et al., 2002) before knowing their safety with great certainty.

To improve the effectiveness of treatment for patients with chronic diseases and comorbidities and the prevention of the emergence of resistance to individual drugs, combination therapy is a new trend in real-world clinical research (Luzuriaga et al., 1997; Mottonen et al., 1999; Wald et al., 2009; Ascierio and Marincola, 2011; Yardley, 2013), which has been effective in the treatment of HIV as well as certain forms of leukemia. Simultaneous combination therapy like two-drug therapy and triple therapy holds huge promise for cancer treatment from the point of cancer cell evolutionary dynamics (Bozic et al., 2013). However, the mechanism and principle underlying effective combination therapy is still a mystery waiting to be investigated (Pritchard et al., 2013).

As a kind of classical combination therapy, traditional Chinese medicine (TCM), which includes multiple herbal ingredients, has been used by Chinese practitioners for thousands of years in real-world clinical settings. Pioneer research (Wang et al., 2008) has already shown that TCM clinical herbal prescription, called formula, consists of both principal and adjuvant components to yield synergy for disease treatment. This demonstrates the molecular mechanism of a well-recognized principle called “*Jun-Chen-Zuo-Shi*” (JCZS) (He et al., 2015; Duan et al., 2018) in TCM formula theories, which illustrates that a well-organized formula should consist of multiple herbal ingredients with differentiated roles like principal role and adjuvant role. Some adjuvant components should be considered to facilitate the delivery of the principal element to the disease site in the body. In real-world clinical herbal prescriptions, some of these have a good therapeutic effect, while others have a poor therapeutic effect. However, the reason why some prescriptions are more effective than others remains to be studied, and there is still a lack of appropriate method to evaluate whether many herbs grouped in certain formulae

are suitable for the treatment of specific disease conditions. Therefore, investigation of herbal combination regularities in herbal prescription will benefit the pharmacological understanding of TCM treatment and discovery of novel combination drugs (Li et al., 2010b). In this study, we proposed a network-based approach to investigate the clinical principle of herbal prescribing and quantify the molecular network interactions between herb target modules of a given clinical prescription in the context of the human protein-protein interaction (PPI) network. In addition, we further investigate the correlation between the organization degree of herbal combinations in clinical prescription and their underlying molecular network interaction closeness, which might partially elucidate the organization principle of JCZS in TCM formula theories.

MATERIALS AND METHODS

Herbal Prescription Dataset

To investigate the regularities of herbal combinations in TCM clinical herbal prescriptions, we collected five different datasets from real-world TCM clinical encounters. These five datasets are described in brief in **Table 1**.

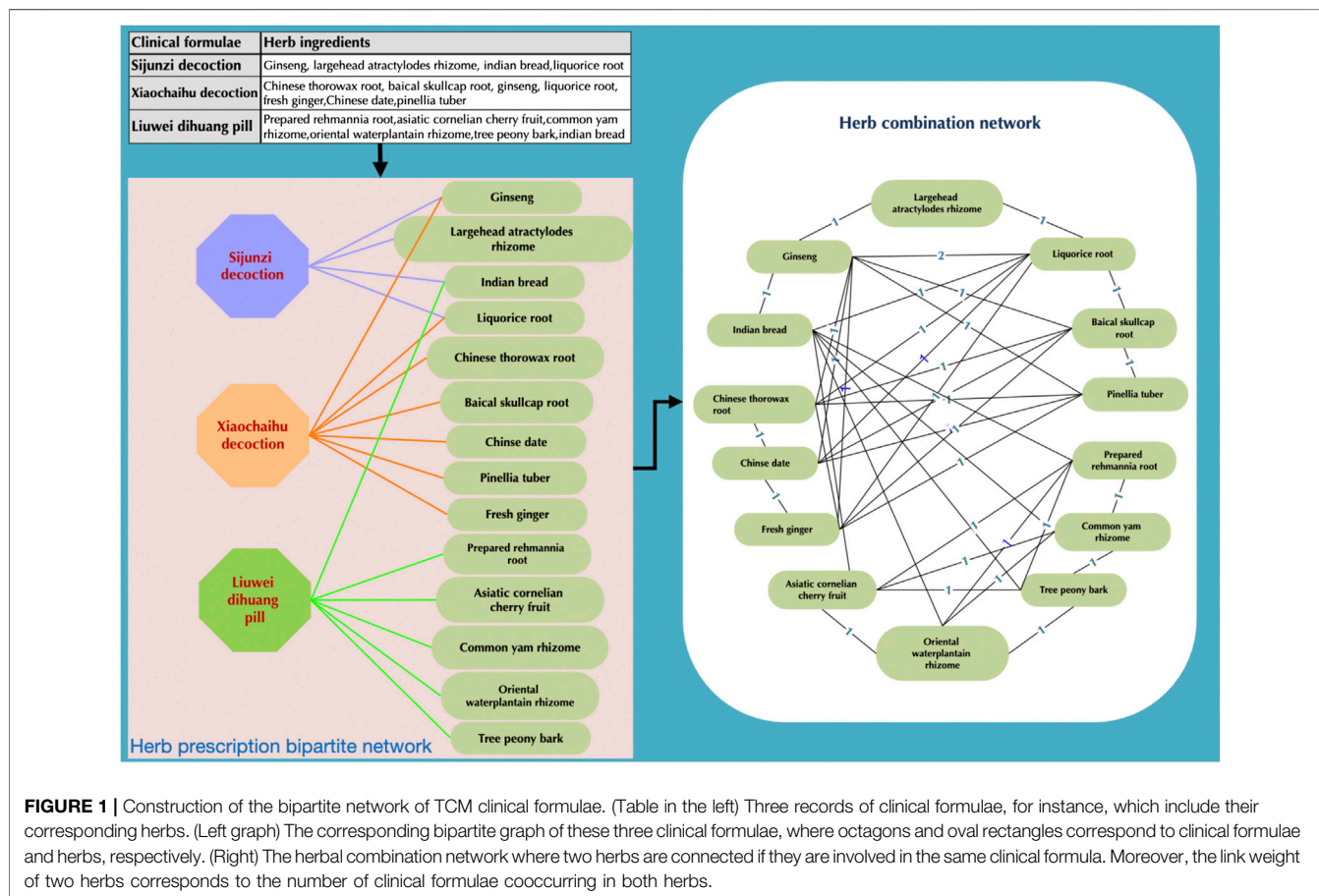
Outpatient formula data was extracted from the outpatient encounters with one and a half years in one well-recognized general TC hospitals in Beijing, China. The data includes 531,284 different clinical formulae that were used by hundreds of TCM physicians for the management of various diseases (e.g., diabetes, CHD (coronary heart disease), stroke, cancer, chronic gastritis, insomnia, and menopause syndrome) in outpatient encounters. The other four datasets are from inpatient clinical cases, which particularly were prescribed for the patients with type 2 diabetes, CHD, qi deficiency, and blood stasis, respectively. The above-mentioned two diseases (i.e., type 2 diabetes and CHD) are major chronic diseases (He et al., 2019) treated by TCM in real-world clinical settings. The other two syndromes, namely, qi deficiency and blood stasis, are two popular TCM diagnoses for various diseases (Wang et al., 2017). These four inpatient datasets are from a TCM clinical data warehouse (Zhou et al., 2010), which collected structured electronic medical record data from real-world clinical settings in about ten top-level TCM hospitals or wards mostly located in Beijing, China. We calculated the distribution of the number of herbs in one formula for each of the five datasets that represent five clinical prescriptions. The average number in each of the five clinical prescription datasets is 13.43 (outpatient), 9.66 (qi deficiency), 11.6 (diabetes), 12.03 (CHD), and 10.13 (blood stasis). We see that most clinical herbal prescriptions have 5–20 different herbs (over 95% of the five clinical formula datasets have 5–20 herbs) and have more than ten herbs averagely, which showed that clinical herbal prescription is a kind of particular multi-ingredients or drug therapy.

Construction of Herbal Combination Network

Considering a clinical formula with multiple herbal ingredients, we can construct a bipartite graph (**Figure 1**) which has clinical formulae and herbs as two types of nodes. The herbal

TABLE 1 | The outline information of five different data sets (in columns) used for analysis.

	Outpatient	Diabetes	CHD	Blood stasis	Qi deficiency
Total number of formulae	531,284	21,626	9,054	2,802	2,393
Distinct number of herbs	576	492	436	439	422
Average frequency of herbs	14,230.02	551.57	270.51	70.65	59.98
Average number of herbs in one formula	15.43	12.55	13.03	11.07	10.58

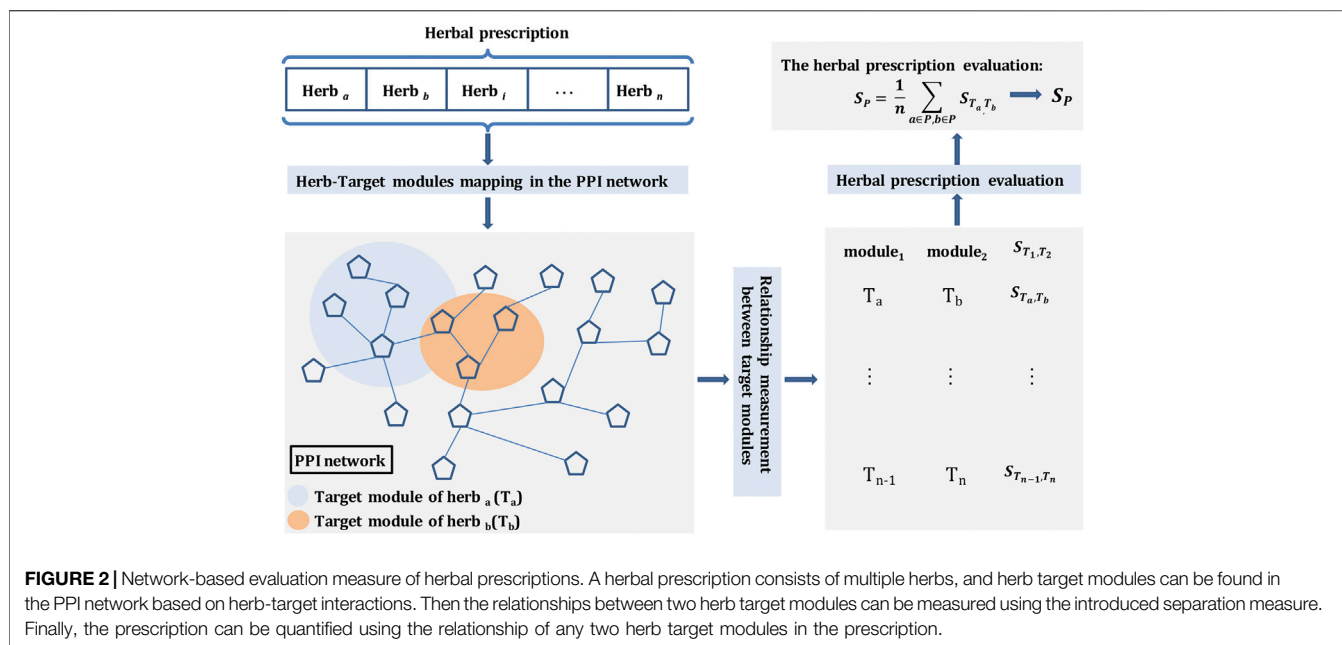


combination network was constructed by considering herb pairs used in one specific clinical formula if they have been prescribed in more than one clinical formula. Then we would generate links (edges) between these herb pairs. This also means that we would generate a complete graph with each herb in one clinical formula. While a large number of clinical formulae are considered, we can generate a herbal combination network (HCNet, see **Figure 1**) with nodes representing herbs (herbal ingredients) and edges with a weight representing prescribing cooccurrence in clinical formulae. If two herb pairs are used more frequently than other herb pairs, they would be having a higher weight than other pairs. Furthermore, to compare the patterns of real-world data to those of randomly generated data, we generated the corresponding simulation data with random permutation by using Fisher

shuffling (Fisher and Yates, 1948) to get random herb ingredients from herb dictionary to a clinical formula.

Network Topological Measurements

To investigate the topological properties of HCNet, we use node degree, clustering coefficient, and link weight distribution measurements, which have been widely used in complex network-related studies. The degree of a node is the number of edges incident to the node, which also means the number of herbs combined with an herb in the HCNet. Clustering coefficient evaluates the link density of the neighborhood of a node, which would be near one if most of the nodes were linked in the neighborhood of a node. We used local clustering coefficient of the undirected network to



calculate for each node in HCNet, which is calculated as follows:

$$C_i = \frac{2|e_{jk} : v_j, v_k \in N_i, e_{jk} \in E|}{k_i(k_i - 1)} \quad (1)$$

in which N_i is the node set of the neighborhood of a node v_i in a graph $G(V, E)$ and k_i is the degree of the node v_i . As a weighted network, HCNet has weight w_{ij} between nodes v_i and v_j , which represents the combination occurrence of herb pairs. We use link weight distribution $P(w)$, which denotes the possibility of a link to have a weight w , to investigate the herbal combination behaviors in HCNet and a given single clinical formula.

Network-Based Measures of Molecular Interactions Among Herbs in Prescriptions

As a kind of classical combination therapy, the herbal prescription consists of multiple herbs to treat clinical diseases. Therefore, we adopted a molecular network-based measurement (Menche et al., 2015; Cheng et al., 2019) to quantify the degree of molecular interactions of herbal combinations in each prescription through calculating the average shortest path length between herb target modules (molecular subnetwork of targets) in the context of human PPI network (Figure 2).

Firstly, we measured the network relationship of herb target modules T_a and T_b in the prescription using the recently introduced separation measure (Menche et al., 2015) to reflect their target localization (Eq. 2):

$$S_{T_a, T_b} = \langle d_{T_a, T_b} \rangle - \frac{\langle d_{T_a, T_a} \rangle + \langle d_{T_b, T_b} \rangle}{2} \quad (2)$$

in which T_a and T_b represent the target modules of herb a and herb b , respectively. S_{T_a, T_b} compares the mean shortest distances

between proteins within each herb, d_{T_a, T_a} and d_{T_b, T_b} , to the shortest distances d_{T_a, T_b} between herb a and herb b target pairs. If proteins are associated with both a and b , $d_{T_a, T_b} = 0$. For $S_{T_a, T_b} < 0$, the targets of the herb a and herb b are located in the same network neighborhood, while for $S_{T_a, T_b} \geq 0$, the two herb targets are topologically separated.

Then, we quantified the herbal prescription using the average of S value between any two herb target modules in the prescription (Eq. 3):

$$S_P = \frac{1}{n} \sum_{a \in P, b \in P} S_{T_a, T_b} \quad (3)$$

in which S_P represents the herbal prescription score and n is the number of herbs in the prescription.

Hierarchical Organization Measure of Clinical Prescriptions

To further explore the associations between the hierarchical property of prescriptions and the molecular network correlations of the herb ingredients in prescriptions. We use entropy to measure the hierarchical organizing degree of a given prescription in terms of the distribution of herbal combinations.

After we obtained the herbal combinations of each prescription and all the prescription groups in our study (i.e., four types of clinical prescriptions), we could record each herbal combination with its weight and compute the distribution of the weight of herbal combinations for each prescription with regard to the global distribution of herbal combinations of each type of clinical prescriptions. Specifically, according to the weight distribution of herbal combinations of a given clinical prescription dataset, we divided the weight range (e.g., from 1

TABLE 2 | The basic information of five different HCNets used for analysis.

	Outpatient	Diabetes	CHD	Blood stasis	Qi deficiency
Total number of nodes	576	492	436	439	422
Total number of edges	120,619	34,824	29,381	19,323	18,186
Average degree in network	419.82	142.56	135.78	89.64	87.39
Edge density	0.73	0.29	0.31	0.20	0.21
Average clustering coefficient	0.85	0.61	0.63	0.51	0.51

to 3,000) into four intervals of some specific value (i.e., 300 in our study). Then using the list of herbal combinations and their weights, we could further calculate the probability of herbal combinations belonging to each interval of the corresponding prescription group for a given prescription. Next, we quantified the hierarchical degree of a prescription based on the entropy measure (Shannon, 1948; Wallace et al., 2019) as follows (Eq. 4). The larger the entropy value, the stronger the hierarchical organization degree for a given prescription, which would medically mean a more systemically organized herbal prescription with hierarchical roles (i.e., tend to prescribe herbal combinations in one formula with diverse global weight ranges in each group). The higher the entropy value, the more the hierarchical organization of the prescription in terms of diverse roles.

$$E = - \sum_{i=1}^n p_i \log p_i \quad (4)$$

where n represents the number of weight intervals and is set to four in our analysis and p_i represents the probability of the herbal combinations in each interval and is calculated as follows (Eq. 5):

$$p_i = \frac{N_i}{\sum_{i=1}^n N_i} \quad (5)$$

in which N_i represents the number of herbal combinations in weight interval i .

Therefore, we could calculate entropy for all prescriptions for each type of the clinical prescriptions. Next, for each type of the clinical prescriptions, we calculate the Pearson Correlations Coefficient (PCC) between its entropy value and S_p score to evaluate the statistical correlation between hierarchical degree and molecular network interactions of the herbal combinations for each prescription group.

RESULTS

Herbal Combination Networks

According to the clinical formulae-herb relationships in outpatient, type 2 diabetes, CHD, qi deficiency, and blood stasis, we constructed five types of herbal combination networks in which nodes represented the herbs and edges represented the cooccurrence of herbs in clinical formulae. These five HCNets were described in brief in Table 2.

Hierarchical Organization of Herbal Combinations in Clinical Prescriptions

To investigate the hierarchical properties of herbal combination networks, we evaluate the relationship between the clustering coefficient and node degree in the network. We performed experiments on four types of clinical prescriptions, and the results were shown in Figure 3.

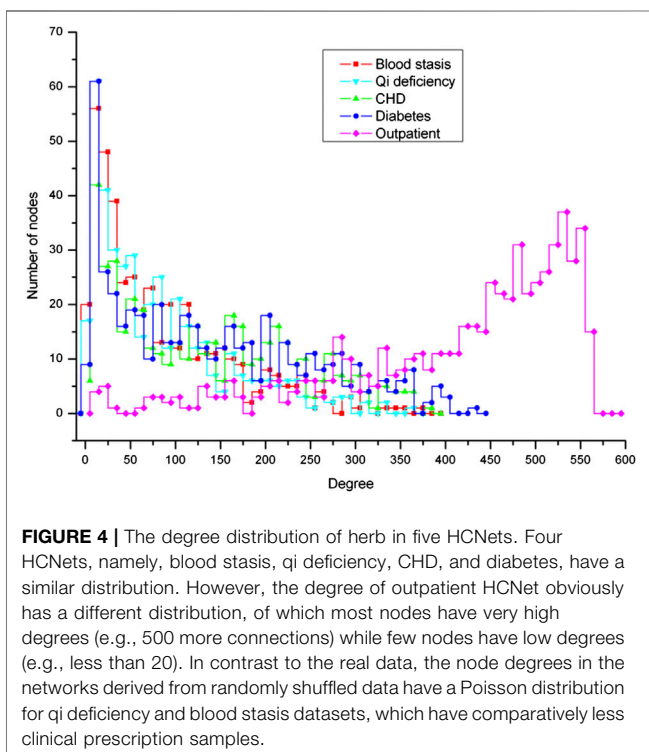
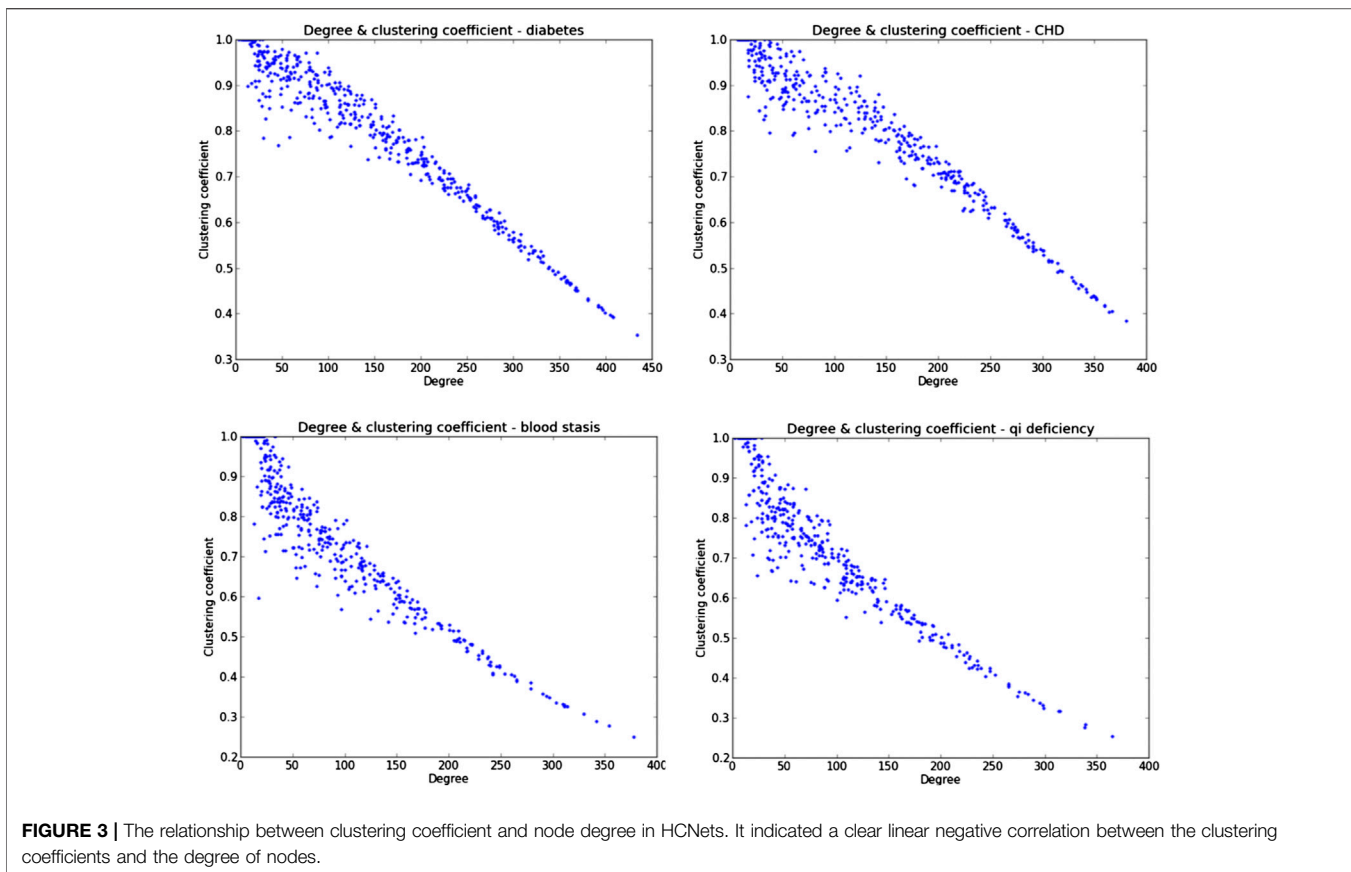
We found that it displays a clear linear negative correlation between the clustering coefficients and the degree of nodes. It was found that, in real networks, clustering coefficients decrease with the vertex degree, which has been taken as a signature of the network hierarchical structure (Soffer and Vazquez, 2005). Therefore, this indicates that hierarchical properties exist in the herbal combination networks. Next, we would further investigate the scaling properties of HCNets according to node degree and weight distribution.

Scaling Heterogeneity of Degree and Weight Distribution

To investigate the scaling properties of HCNets, we evaluate the node degree distribution and link weight distribution in the network. It showed that herbal combinations in clinical formulae are obeying a scaling property, which has significantly departed from Poisson distribution (Figure 4). Although the node degree distribution in HCNets is not power law (this is obviously rational because the capacity of herb dictionary is fixed and thus increase of nodes is not met in HCNets), it showed a significant heterogeneous scaling for different nodes. The degrees of herb nodes scale from less than ten (e.g., 5) to several hundred (e.g., 550), whose range is much wider than the corresponding Poisson distribution.

Furthermore, it is interesting that the link weight distribution of HCNets is a clear power law, in which several herbal combinations are prescribed in high frequency while most of the other herbal combinations have very low frequency (Figure 5). This means that, in clinical practice, the prescription of herbal combinations for treatment is nonrandomly chosen and preferentially used by TCM physicians for the disease management of individualized patients.

We find that herbal combinations in four clinical formulae are obeying a scaling property (Figure 5), and it also means that the distribution of herbal combination has a hierarchical property. Next, we will calculate the degree of interaction between the targets in the context molecular network in herbal combinations and investigate whether frequently used herbal combinations hold close pharmacological effects.



Molecular Network Interaction Patterns of Herbal Combinations

To evaluate the underlying pharmacological mechanisms of herbal combinations in clinical prescriptions, we quantified the network correlations between the targets of the herbal ingredients of the four types of clinical prescriptions for diabetes, CHD, qi deficiency, and blood stasis (see Methods section, *Network-Based Measures of Molecular Interactions Among Herbs in Prescriptions*).

We collected the protein targets of herbs in prescriptions from HIT database (Ye et al., 2011), which is a comprehensive and fully curated database of herbs and their corresponding protein targets. Due to the lack of target information for some herbs according to HIT database, at the same time, to assure each prescription should contain at least two herbs in the analysis. Finally, we quantified the molecular network correlations of those herbs in 21,566, 9,051, 2,392, and 2,801 prescriptions from diabetes, CHD, qi deficiency, and blood stasis, respectively, in which the number of herbs containing known targets is greater than or equal to two. The mean and standard deviation (std) of S_p scores of four types of prescriptions were shown in **Table 3**.

In order to give a benchmark reference for the four types of clinical prescriptions, we selected the classical prescriptions of TCM, Treatise on Exogenous Febrile Disease (TEFD), for comparative experiments. The TEFD contained 113 classical prescriptions in total, of which 85

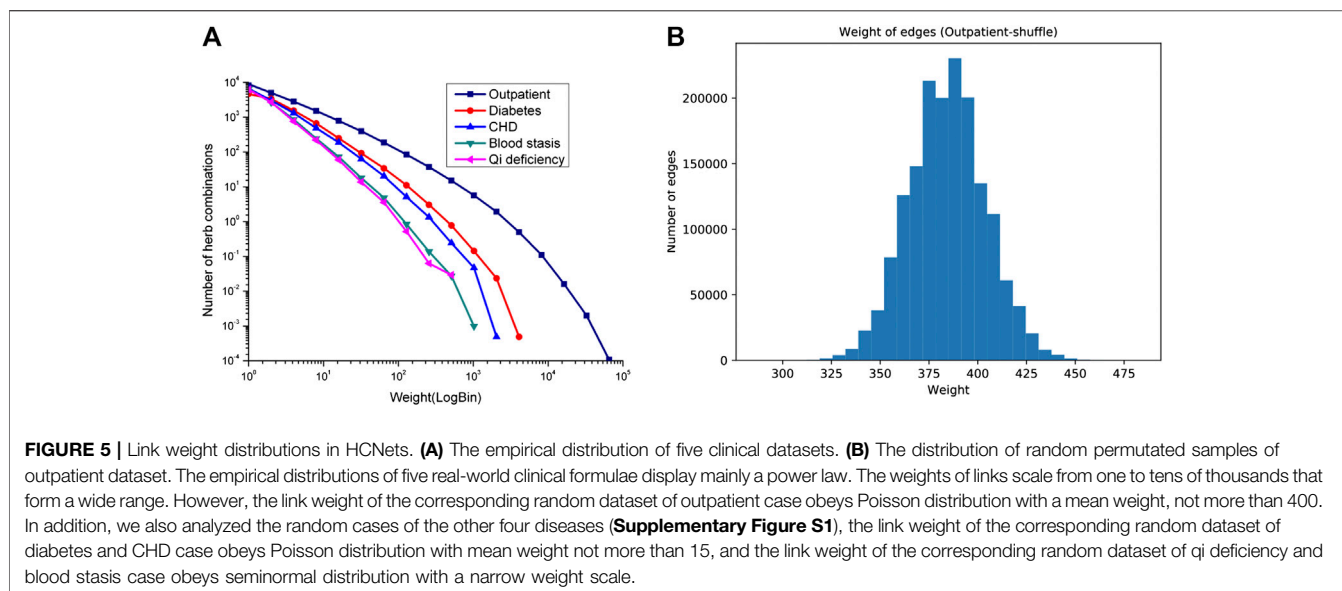


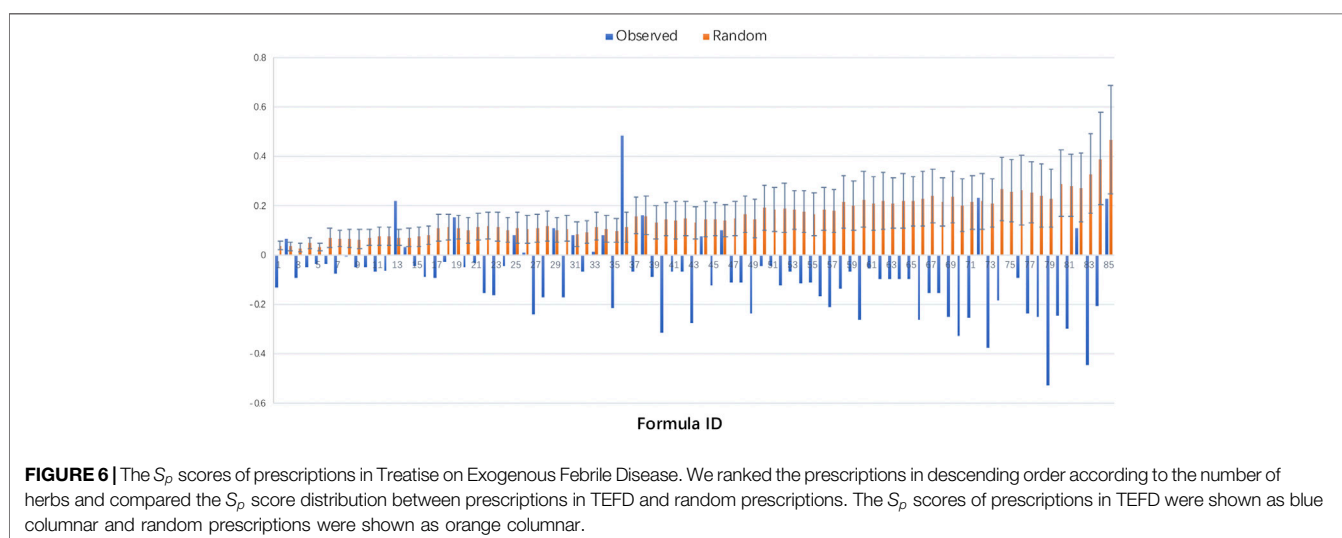
TABLE 3 | The mean and standard deviation of S_p scores of four types of clinical prescriptions and TEFD.

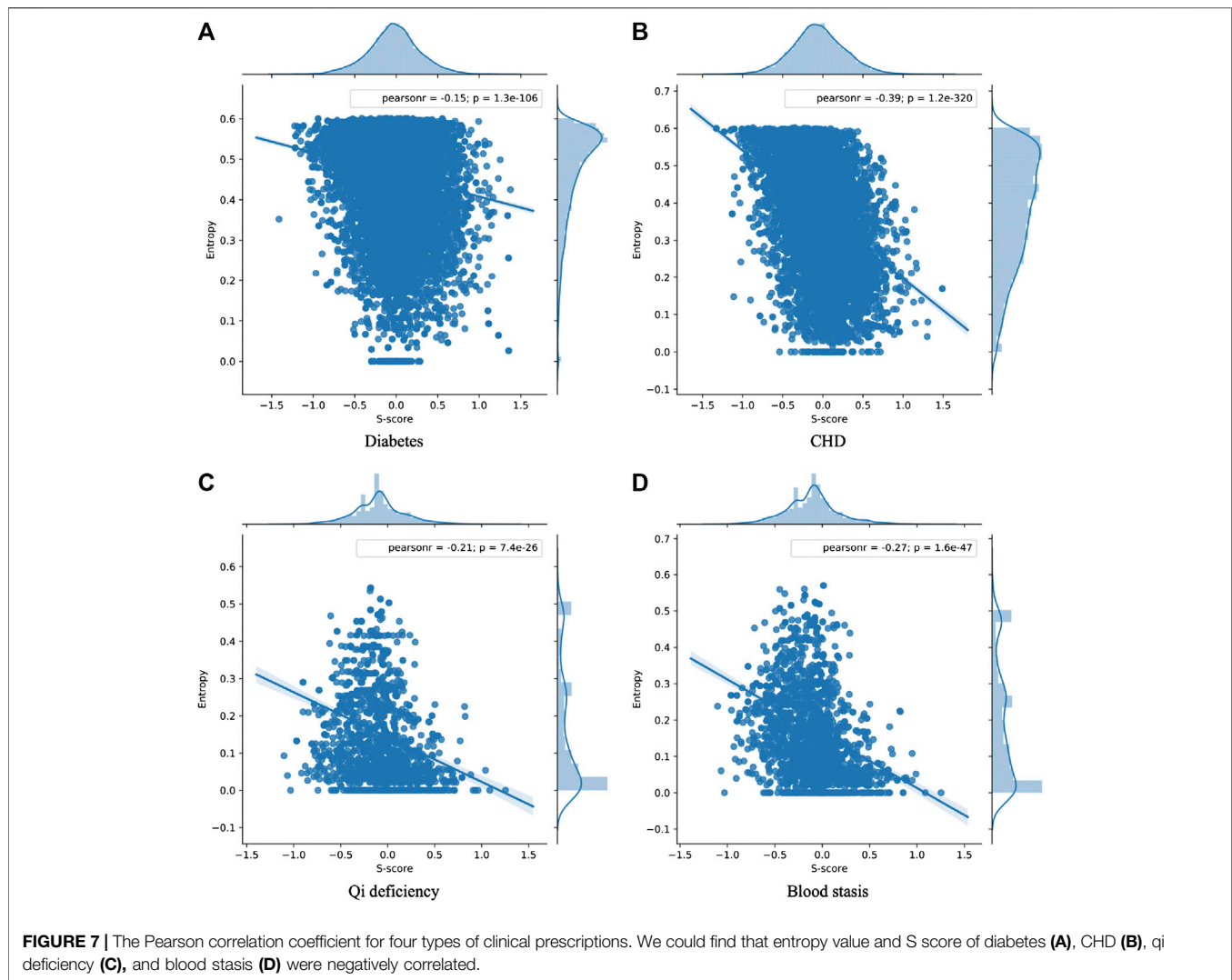
Type	Diabetes	CHD	Qi deficiency	Blood stasis	TEFD	Random
Mean	-0.03	-0.05	-0.09	-0.11	-0.09	0.16
Std	0.32	0.33	0.28	0.28	0.15	0.08

prescriptions with associated targets were selected for quantification. The number of herbs containing known targets is greater than or equal to two in 85 prescriptions. We collected the classical prescriptions from TEFD as a representative prescription benchmark with high-quality herbal combinations because most prescriptions in TEFD have been widely used in TCM clinical practice for thousands of years (Li et al., 2010a; Wang and Xiao, 2010; Zhang et al., 2010). Therefore, in

general, we will consider that the herbal combinations held in the prescriptions from TEFD give a better example than those from the above four types of clinical prescriptions.

Next, we quantified the degree of interactions between the herbal combinations in 85 prescriptions from TEFD in the context of the human PPI network (**Figure 6**). For comparison of random expectations, we reshuffled (1,000 random permutations) the herbs in each prescription by using the Fisher-Yates method (Fisher and Yates, 1948). Finally, we found that the S_p scores of prescriptions in TEFD were significantly lower than those of random prescriptions ($p = 2.46718e-27$). We found that 92% of real prescriptions in TEFD had lower S_p scores compared to random prescriptions, and S_p scores of 79% of real prescriptions were less than 0, but S_p scores of all random prescriptions were greater than 0. And we found that the mean S_p score in TEFD was -0.09, but the random was 0.16, indicating that the target modules of herbs in a prescription were closely related





to each other in the context of the molecular network. To further evaluate the quality of herbal combinations of current clinical prescriptions, with similar calculation methods, we obtained the S_p scores of each prescription in the four clinical prescriptions datasets. The results showed that the mean of S_p scores of all four clinical prescriptions was less than 0. This means that most real-world clinical prescriptions would tend to have a high degree of herbal combinations as good as those of TEFD prescriptions. It is practical and reasonable that, in TCM clinical settings, most practitioners would directly adopt the classical prescriptions or use the classical prescriptions as a basic therapeutic framework for clinical treatments (Duan et al., 2018; Chen et al., 2019).

Molecular Correlations of Hierarchical Herbal Combinations in Clinical Prescriptions

We have found that the weights of herbal combinations scale from one to tens of thousands that form a wide range and the distribution of herbal combinations has a hierarchical

organization property in our previous analysis. Meanwhile, we also obtained the molecular measures of herbal combinations in prescriptions in terms of their molecular interactions. Next, we would expect the potential associations between these two indexes for a given prescription (see Methods section, *Hierarchical Organization Measure of Clinical Prescriptions*).

To test the assumption, we performed a Pearson correlation analysis on its entropy value and S_p score. The PCC and p -value were shown in **Figure 7**. We could find that entropy value and S_p score of clinical prescriptions were negatively correlated with PCC ranging from -0.15 to -0.39 (**Figure 7A**, $p = 1.3e-106$; **Figure 7B**, $p = 1.2e-320$; **Figure 7C**, $p = 7.4e-26$; **Figure 7D**, $p = 1.6e-47$) for the four clinical herbal prescriptions. This indicated that the larger the entropy value of a given prescription, the smaller the S_p score of the prescription. Therefore, it indicated that the more the hierarchical herbal combinations were involved in a prescription, the more the connected molecular networks would be found between the targets of the herbal ingredients in a given prescription. The results also reflected exactly a well-recognized principle called JCZS in TCM formula theories,

which illustrates that a well-organized formula should consist of multiple herbal ingredients with differentiated therapeutic roles for targeted disease conditions.

DISCUSSION AND CONCLUSION

TCM herbal prescription which usually includes tens of different herbal ingredients as a single therapeutic formula is the main kind of intervention for chronic disease treatment in China (Qi et al., 2013). Typically, it is a kind of combination therapies with complicated underlying molecular pharmacological mechanisms (Chan et al., 2012). Combination therapies offer promising therapeutic solutions for complex disease treatments, from hypertension, COPD, and cancer with potential high efficacies and lower adverse effects (Yki-Jarvinen, 2001; Margulies and Hicks, 2009). However, the clinical organization principle and its underlying network mechanisms still need to be elucidated. TCM herbal prescription has been widely used as a combination therapy in clinical settings (He et al., 2012; Liu et al., 2013; Duan et al., 2018) by adhering to empirical clinical regularities, such as JCZS (He et al., 2015; Duan et al., 2018), and frequently used herbal combinations. Connecting those clinical regularities with the molecular network patterns related to the targets of herbal ingredients would potentially improve the pharmacological understanding of the empirical rules to develop more effective combination therapies.

The well-recognized principle called JCZS in TCM formula theories means that a well-organized formula should consist of multiple herbal ingredients with different therapeutic roles (Wang et al., 2008). This would imply the existence of a hierarchical organization of herbal combinations and the underlying network mechanisms of herbal ingredients for a well-established set of clinical prescriptions (e.g., the herbal prescriptions in TEFD). Here, we investigated the clinical regularities of herbal combinations involved in each prescription in terms of network topological patterns and applied a network-based measurement (Cheng et al., 2019) to quantify the molecular network interactions between herb target modules in a given herbal prescription. Our results indicate that there do exist clinical prescription rules to choose herbal combinations in a clinical prescription, which can be displayed by the scaling and hierarchical topological properties of the herbal combination network. Specifically, a clinical prescription would tend to consist of both highly used herbs or herbal combinations and less used herbs or herbal combinations as a whole formula for disease treatment. This might resemble to the well-established prescription principle (i.e., JCZS) for TCM herbal prescriptions, which implies a good prescription need include herbs with different pharmacological effects. Furthermore, we found that the network-based measure of high-quality drug combinations (Cheng et al., 2019) could be used for evaluating the molecular interaction closeness of both herbal combinations and prescriptions. For example, a classical formula in TEFD, “Shaoyao Gancao decoction” (SGD), contains two herbs: Radix Paeoniae Alba (hRPA) and Radix Glycyrrhizae Preparata (hRGP), which has been further prescribed as core herbal combinations in many classical formulae [e.g., Mahuang Shengma decoction (Fan et al., 2011), Xiaoqinglong decoction (Wang et al., 2018), Gegen

decoction (Chai et al., 2020), Chaihu Guizhi decoction (Li et al., 2019), and HuangQin decoction (Dai et al., 2018)]. According to the molecular network-based measurement, we found that the S_p score of SGD was -0.03 (the random expectation was 0.03). This means that close interactions are involved between the target modules of the two-member herbs in SGD. Actually, by identifying the herb-target associations from HIT database, we found that there are 47 and 24 potential targets of hRPA and hRGP, respectively. These two target sets each form a rather dense PPI subnetwork (we call it a module, **Figure 8A**). There are eight common targets shared by hRPA and hRGP, such as TNF, CASP3, RELA, and NOS2. Meanwhile, 75 interactions exist between the target modules of hRPA and hRGP (with 0.113 network density). We further used DAVID (Huang Da et al., 2009) to obtain the enriched GO and KEGG pathways (**Figure 8B**). The results showed that SGD enriched the inflammation-related pathways such as Hepatitis B, NF- κ B, and TLRs (Mantovani, 2010) and liver-related GO annotations such as liver regeneration, which could partially explain the clinical effects of SGD for treating liver disorders and anti-inflammatory in clinic settings (Bi et al., 2014).

Overall, we have been working to investigate the network topological regularities and the underlying molecular network mechanisms of herbal prescriptions, and we hope that our work and findings for clinical prescriptions would give references for understanding of the principles and molecular mechanisms of multidrug combinations (Dawson and Carragher, 2014; Cheng et al., 2019; Zhang et al., 2019) after fully digging the hidden knowledge underlying the effective multiple therapies (e.g., Chinese herbal prescriptions).

This study has several limitations. First, since it is not aiming to predict the specific knowledge on pharmacological effects of specific prescriptions or drugs, unlike the study on pharmacological research of a given herbal prescription or predicting a novel target, it would be difficult for us to design a straightforward wet-lab experiment to validate the rationality of organizing herb combinations in a good clinical prescription. However, large-scale wet-lab experiments on validating the differentiated molecular patterns between high frequently used herbal combinations and rarely used herbal combinations should be designed and performed in further research to improve the reliability of the network analysis results. Second, currently, we only used HIT database as the herb-target associations for molecular network-related analysis, which would limit the number of herb-target associations and thus influence the generalization of our analysis. We would integrate other well-established network pharmacological databases, such as ETCM (Xu et al., 2019) and SymMap (Wu et al., 2019) to further validate the underlying molecular network regularities of herbal prescriptions. Third, due to the complexity of clinical conditions of individualized patients, it is important that the prescribed herbal treatments for specific patients should be evaluated by the outcomes and finally help figure out the true high-quality clinical herbal prescriptions from a large-scale clinical dataset. Therefore, we would extend the evaluation measure together with disease conditions to find good herbal prescriptions with potential effectiveness. However, without the related in vitro or in vivo experimental validations, we believe that

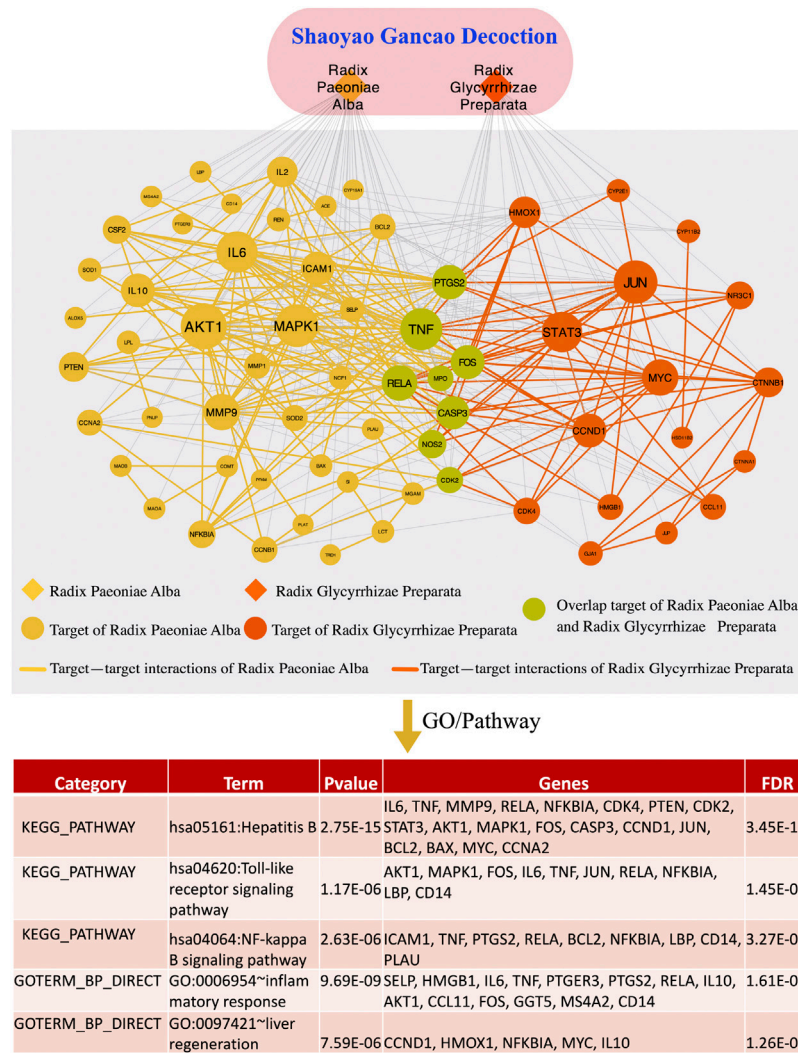


FIGURE 8 | The network molecular associations for the only two herbs, namely, Radix Paeoniae Alba and Radix Glycyrrhizae Preparata in the Shaoyao Gancào decoction, (SGD). **(A)** Target modules of the two herbs and their interactions. Yellow nodes represent the targets of Radix Paeoniae Alba, and red nodes represent the targets of Radix Glycyrrhizae Preparata, while the edges represent the interactions between the targets in the PPI network. Nodes with green color represent overlapping targets of two herbs. **(B)** The related gene ontology and pathways of target modules of the two herbs.

our current research could deliver certain potential insights for the field of network pharmacology and combination drug development. By evaluating the underlying molecular network patterns of the herbal combinations in herbal prescriptions, it would be promising to propose a novel network pharmacological approach to investigate the inherent network mechanisms (e.g., organizing of herbal combinations) of a given clinical prescription and thus help design combination therapies with better effectiveness for disease treatment.

DATA AVAILABILITY STATEMENT

All datasets presented in this study are included in the article/Supplementary Material.

AUTHOR CONTRIBUTIONS

XZ, ZG, RZ, and BL conceived and designed the study. NW and ND analyzed data and wrote the draft manuscript; KY, ZS, and KC collected related data; YP, RZ, DW, JY, CJ, YZ, and XL reviewed the methods and the results. All authors have reviewed the manuscript; in particular, XZ and DW have revised the manuscript with the help of other coauthors.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fphar.2020.590824/full#supplementary-material>.

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Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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