Understanding the diverse functions of Huatan Tongluo Fang on rheumatoid arthritis from a pharmacological perspective

CHUNSONG ZHENG[1], MINGSHAN QIU[2], XIAOJIE XU[1,3], HONGZHI YE[1,4], QIAN ZHANG[2], YIHAN LI[2], XIANXIANG LIU[1] and JINCHUN CHEN[2]

[1]Institute of Bone Disease, Academy of Integrative Medicine, Fujian University of Traditional Chinese Medicine, Fuzhou, Fujian 350122; [2]Department of Rheumatism and Immunology, Affiliated Xiamen Hospital of Traditional Chinese Medicine, Fujian University of Traditional Chinese Medicine, Xiamen, Fujian 361009; [3]College of Chemistry and Molecular Engineering, Peking University, Beijing 100871; [4]Fujian Key Laboratory of Integrative Medicine on Geriatrics, Fujian University of Traditional Chinese Medicine, Fuzhou, Fujian 350122, P.R. China

Received January 26, 2015; Accepted March 3, 2016

DOI: 10.3892/etm.2016.3329

Abstract. Huatan Tongluo Fang (HTTLF) is a traditional herbal formula that can resolve phlegm and dredge collaterals. HTTLF has also been used to treat rheumatoid arthritis (RA); however, the mechanism underlying the therapeutic effects of HTTLF on RA has not been clearly elucidated at the molecular level. In the present study, an integrated model of system pharmacology containing chemical space analysis, potential active compound prediction and compound-target-disease network was constructed to investigate the molecular mechanisms of HTTLF. The compounds from HTTLF dispersed well in the chemical space. Most of the compounds from HTTLF had similar chemical spaces to drug/drug-like compounds associated with RA, according to the MDL Drug Data Report. A total of 127 potentially active compounds and 17 targets of RA were identified. Among them, 50 compounds interacted with ≥2 targets, while 77 compounds interacted with only one target. In addition, 17 targets were associated with 82 diseases that belonged to 26 categories. These results indicate that HTTLF has diverse chemical spaces and polypharmacology with regards to the treatment of RA. In addition, HTTLF demonstrated therapeutic potential against diverse diseases other than RA, including osteoarthritis, atherosclerosis and brain cancer. This study provides a novel platform for understanding how HTTLF treats RA; this is beneficial for explaining the diverse functions of HTTLF with regards to RA, and may help develop novel compounds with desirable therapeutic targets to treat RA.

Correspondence to: Professor Jinchun Chen, Department of Rheumatism and Immunology, Affiliated Xiamen Hospital of Traditional Chinese Medicine, Fujian University of Traditional Chinese Medicine, 1739 Xianyue Road, Xiamen, Fujian 361009, P.R. China
E-mail: chenjinchunfjtcm@163.com

Abbreviations: RA, rheumatoid arthritis; TCMs, traditional Chinese medicines; HTTLF, Huatan Tongluo Fang; BA, *Bile arisaema*; SP, *Semen persicae*; FC, *Flos carthami*; SA, *Sinapis alba*; BB, *Bombyx batryticatus*; PA, *Paeonia alba*; CIA, collagen-induced arthritis; VEGF, vascular endothelial growth factor; MMP, matrix metalloproteinase; MMFF, Merck molecular force field; MW, molecular weight; nHDon, number of hydrogen bond donors; nHAcc, number of hydrogen bond acceptors; AlogP, octanol-water partition coefficients; nRB, number of rotatable bonds; MPSA, molecular polar surface area; PDB, Protein Data Bank; COX-2, cyclooxygenase-2; JAKs, janus kinases; TNF-α, tumor necrosis factor α; Syk, spleen tyrosine kinase; MAP, mitogen-activated protein; Std Dev, standard deviation; TTD, Therapeutic Targets Database; QSAR, quantitative structure-activity relationship

Key words: rheumatoid arthritis, systems pharmacology, Huatan Tongluo Fang, traditional Chinese medicine, diverse functions

Introduction

Rheumatoid arthritis (RA) is an autoimmune disease primarily characterized by arthrosynovitis (1). Its main clinical manifestations are chronic, symmetrical, multi-joint synovitis and articular damage (2). The incidence of RA is ~1% worldwide; RA severely influences quality of life and health as it can result in a high level of disability, and negatively affects individuals, families and society (3). At present, there is no individually recognized drug to control and treat RA. The primary drugs on the market for RA treatment are non-steroidal anti-inflammatory drugs, biological agents, disease-modifying anti-rheumatic drugs and glucocorticoids. However, the pharmacological management of RA has targeted the symptoms of the disease, rather than the underlying causes(4). In addition, the prolonged use of these drugs has numerous side-effects, and they are becoming ineffective as a result of drug resistance (5). Thus, it is important that researchers develop novel anti-rheumatic drugs that delay the progression of RA and reduce disability.

Traditional Chinese medicines (TCMs) have been used to treat RA for >2,000 years. It has been demonstrated that

the effect of herbal formulas on RA is an integrated result of various mechanisms of action, including immunity adjustment and inflammatory control (6). Herbal formulae serve a moderate role in the treatment of RA; they have few side-effects and are suitable for long-term use (7,8). The herbal treatment of RA has received increasing attention (9); it is thought that they have great potential to be developed and utilized for the treatment of patients with RA.

Huatan Tongluo Fang (HTTLF) is a traditional herbal formula that has been widely prescribed to treat RA in the Xiamen Hospital of Traditional Chinese Medicine (Xiamen, China). HTTLF is composed of six herbs, including *Bile arisaema* (BA; Dannanxing), *Semen persicae* (SP; Taoren), *Flos carthami* (FC; Honghua), *Sinapis alba* (SA; Baijiezi), *Bombyx batryticatus* (BB; Jiangcan) and *Paeonia alba* (PA; Baishao). Clinical observations have demonstrated that HTTLF can reduce the level of vascular endothelial growth factor (VEGF) in the serum of patients with RA, and significantly alleviate the indexes of erythrocyte rate, C-reactive protein, tenderness and swelling of the joints of patients with RA (10). The results of animal model experiments have demonstrated that HTTLF can relieve inflammation in rats with collagen-induced arthritis, and significantly reduce the expression levels of serum VEGF and matrix metalloproteinase (MMP)-3 (11). However, the underlying molecular mechanisms of HTTLF remain unknown. Fortunately, numerous computer simulation methods have made a significant contribution towards understanding the theory of TCMs and their mechanisms of action at a molecular and systems level (12-14). In the present study, an integrated model of systems pharmacology, developed in a previous study (12,13), that combined molecular database building, chemical space, molecular docking and network pharmacological techniques, was used to investigate the molecular characteristics of HTTLF and map a compound-target-disease network to understand the interaction between HTTLF and therapeutic targets of RA from a systematic point of view. These attempts may offer novel opportunities to investigate the pharmacological basis of HTTLF, and provide an effective method to aid development of treatments for RA using herbal formulae.

Materials and methods

Molecular database building. All chemical ingredients from the six herbs of HTTLF were collected from the Chinese Herbal Drug Database, the Handbook of the Constituents in Chinese Herb Original Plants and other literature (15-19). A total of 692 compounds were obtained, of which 144 were obtained from Dannanxing, 68 from Taoren, 163 from Honghua, 119 from Baijiezi, 93 from Jiangcan and 105 from Baoshao. The chemical structures were drawn using ISIS Draw version 2.5 (MDL Information Systems, Inc., San Leandro, CA, USA) and further optimized by Discovery Studio version 2.0 (DS 2.0; Accelrys, Ltd., San Diego, CA, USA) with a Merck molecular force field (MMFF). In addition, 1,362 RA-associated drug/drug-like compounds were collected from the MDL Drug Data Report (20); these were optimized with the MMFF and saved to files in standard definition format in preparation for the subsequent analyses (21).

Table I. 17 key protein targets associated with rheumatoid arthritis.

Protein name	PDB code
Dihydroorotate dehydrogenase, mitochondrial	3O8A
Cyclooxygenase-2	3MQE
Tyrosine-protein kinase Janus kinase 3	3LXL
Tumor necrosis factor-α	2AZ5
Interleukin 1 receptor	1IRA
Integrin α-4	3V4V
Thioredoxin reductase, cytoplasmic	4B1B
Interleukin-2	1M48
Cathepsin K	1AU0
Janus kinase 1	4IVD
Tyrosine-protein kinase SYK	4FZ6
Mitogen-activated kinase p38	1CM8
Metalloproteinase domain-17	2A8H
Inhibitor of nuclear factor κB kinase	3RZF
Matrix metalloproteinase-9	1GKC
Vascular endothelial growth factor receptor 2	1Y6A
Macrophage migration inhibitory factor	4F2K

PDB, Protein Data Bank; SYK, spleen tyrosine kinase.

Chemical space analysis. In the current study, a total of 150 physicochemical properties were calculated by the quantitative structure-activity relationship (QSAR) module of DS 2.0 (13), and principal components analysis was used to map the distributions of HTTLF and drug/drug-like compounds in the chemical space in two dimensions. According to Lipinski's rule of five (22), four important pharmacology-associated descriptors, including molecular weight (MW), the number of hydrogen bond donors (nHDon), the number of hydrogen bond acceptors (nHAcc) and octanol-water partition coefficients (AlogP), were calculated in order to evaluate the drug-likeness of HTTLF compounds.

Molecular docking. To determine whether HTTLF can interact with 17 key targets associated with RA (23,24), molecular docking simulations were performed between HTTLF compounds and these targets by the LigandFit module of DS 2.0. Their protein crystal structures were retrieved from the Protein Data Bank (PDB; Table I) (25). All crystallographic waters were removed from the file and the hydrogen atoms were added. An inhibitor from the PDB file was used to define the active site. HTTLF compounds were docked onto the protein models. The interactions between these were evaluated using DockScore (26). The compounds with the top 20 DockScores were selected as potentially active compounds of HTTLF (13).

Network construction and analysis. Potentially active compounds and corresponding targets were analyzed using compound-target network (CTN) and target-disease network (TDN) (18). The CTN was constructed by linking the potential active compounds and their corresponding targets, and the TDN was constructed by linking the potential targets

Table II. Statistics of key molecular properties of compounds in Huatan Tongluo Fang.

Name	Mean	Standard Deviation	Minimum	Maximum
Carbon count	15.91	10.77	1.00	75.00
Nitrogen count	0.38	0.92	0.00	5.00
Oxygen count	3.50	5.53	0.00	47.00
Octanol-water partition coefficients	3.36	4.16	-4.62	18.62
Molecular weight	278.85	203.10	53.06	1707.20
Number of rotatable bonds	6.97	7.18	0.00	37.00
Number of hydrogen bond acceptors	3.74	5.50	0.00	47.00
Number of hydrogen bond donors	1.97	3.40	0.00	28.00
Molecular volume	199.67	133.85	19.20	985.43
Molecular surface area	292.18	187.51	72.33	1479.01
Molecular polar surface area	66.23	94.03	0.00	812.37
JX	2.54	0.68	0.97	4.70
JY	2.62	0.70	1.02	4.72
Wiener	2301.02	7270.50	9.00	88225.00
Zagreb	94.71	82.67	10.00	682.00

and their corresponding diseases. The associations between the targets and diseases were retrieved from the Therapeutic Targets Database (23). The above networks were generated and analyzed by Cytoscape version 2.8.3 (University of California, San Diego, CA, USA) (27).

Results

Molecular physicochemical property analysis of HTTLF. The distribution of the physicochemical properties of compounds from HTTLF was diverse (Table II). The majority of the compounds were observed to have clustered on the left side of the chemical space (Fig. 1). There was a large overlap between HTTLF compounds and drug/drug-like compounds in the chemical space. Fig. 2 presents the percentages of MW (<500), AlogP (<5), nHDon (<5) and nHAcc (<10) were 95.59, 67.92, 90.89 and 89.31%, respectively. According to the structure-activity relationship theory and Lipinski's rule of five (22,28), the compounds from HTTLF possessed molecular diversity and drug-likeness.

Diverse functions of HTTLF. Docking results demonstrated that there were 127 potentially active compounds in HTTLF. The interactions between compounds and targets are presented in the compound-target network (CTN) (Fig. 3). The CNT showed that 50 compounds can interact with ≥2 targets, while 77 compounds can interact with only one target (Fig. 4). The general network properties and key compounds in the CTN are listed in Tables III and IV, respectively. The values of network heterogeneity and network centralization were 1.336 and 0.108, respectively. In addition, a TDN was constructed (Fig. 5). The TDN demonstrated that 17 targets were associated with 82 diseases (Table V) that belonged to 26 categories, such as musculoskeletal, immune system and cardiovascular diseases, and neoplasms. This suggests that HTTLF may demonstrate efficacy in targeting these diseases.

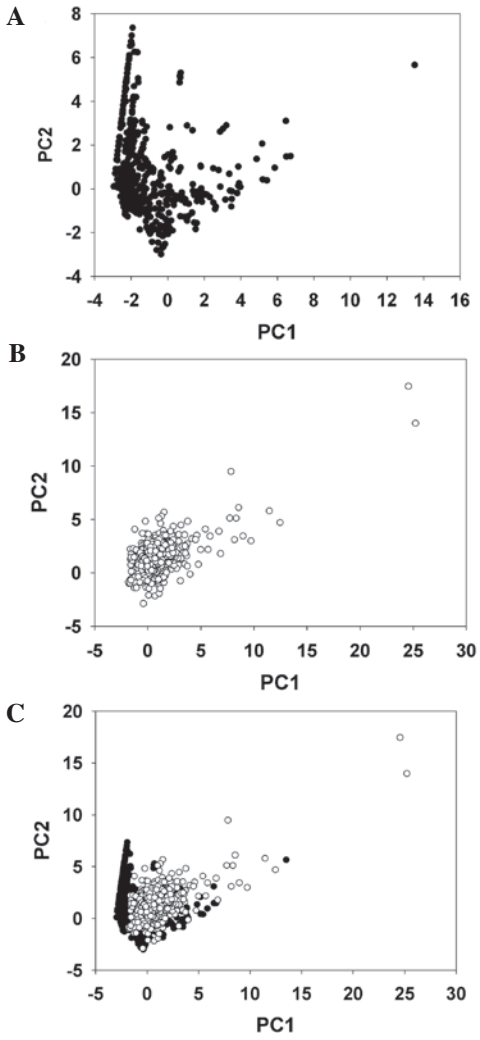

Figure 1. Chemical space distributions of compounds from HTTLF and the MDDR. (A) Chemical space distributions of compounds from HTTLF. (B) Chemical space distributions of compounds from MDDR. (C) Black circles represent compounds from HTTLF, whereas white circles represent drug/drug-like compounds from the MDDR. HTTLF, Huatan Tongluo Fang; PC, principal component; MDDR, MDL Drug Data Report.

Table III. General network properties of the compound-target network.

Parameter	Compound-target network
Number of nodes	144
Number of edges	340
Network density	0.033
Network heterogeneity	1.336
Isolated nodes	0
Number of self-loops	0
Multi-edge node pairs	0
Network centralization	0.108
Shortest paths	20,592 (100%)
Characteristic path length	3.891
Average number of neighbors	4.722

Table IV. Key compounds with top-10 degree in the compound-target network.

Index	Degree	Chemical name	Known
SP-27	12	Naringenin 7-O-beta-D-glucoside	No
BA-75	12	Isoschaftoside	No
BA-74	12	Schaftoside	No
BA-143	12	Apigenin 6, 8-di-C-glucoside	No
SP-68	11	Chlorogenic acid	Yes
SP-31	10	Quercitrin	No
SP-30	10	Quercetin	Yes
SP-1	10	Amygdalin	Yes
BA-54	10	Deacetylcentapicrin	No
BA-144	10	Apioside	No

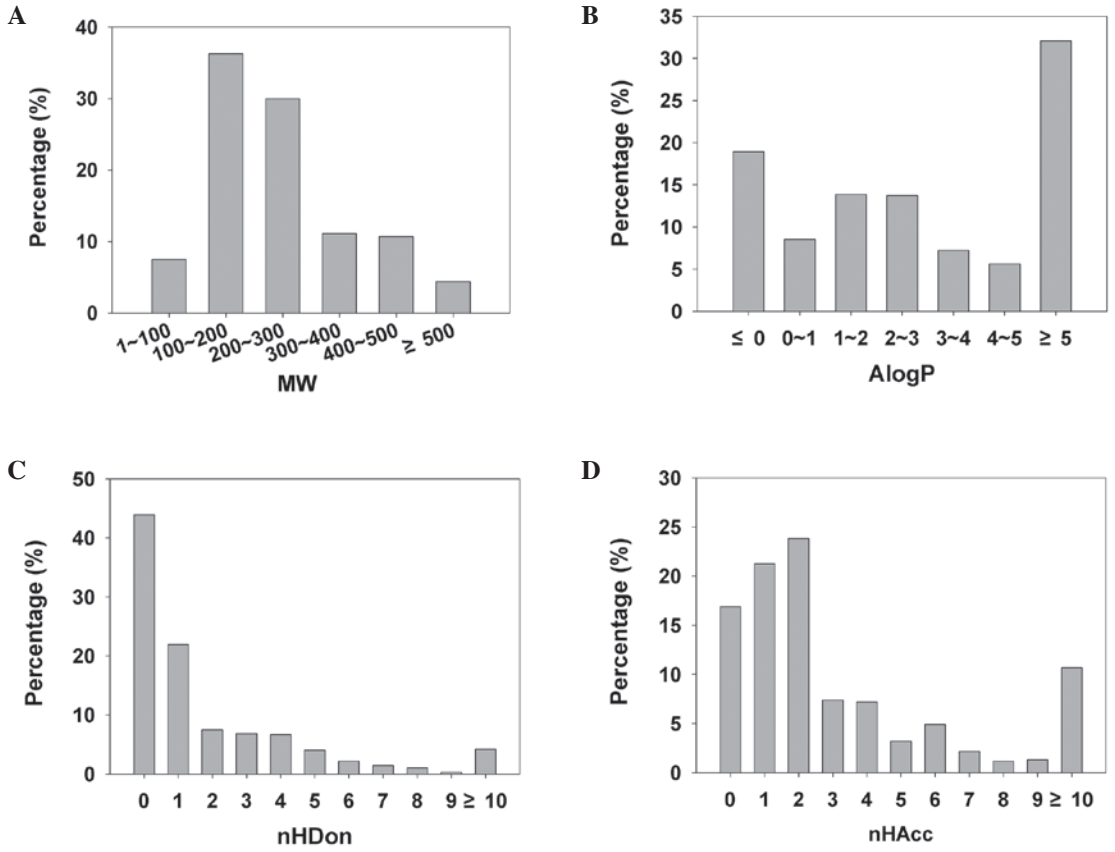

Figure 2. The distributions of four important molecular properties of compounds from Huatan Tongluo Fang. Molecular properties consist of (A) MW, (B) AlogP, (C) nHDon and (D) nHAcc. MW, molecular weight; AlogP, octanol-water partition coefficients; nHDon, number of hydrogen bond donors; nHAcc, number of hydrogen bond acceptors.

Discussion

RA is a common chronic inflammatory disease that results in a considerable burden for the patient and society. The cause of RA is not a single effect; it is caused by multiple molecular abnormalities (29). Numerous clinical studies have demonstrated that a number of Chinese herbal monomers, such as triptolide and sinomenine, have efficient therapeutic effects in treating RA (30,31). It has been demonstrated that a combination of *Tripterygium wilfordii* polyglycoside and methotrexate can improve the therapeutic effects; in addition, the combination treatment can reduce side-effects and drug resistance (32). Furthermore, *Tripterginum wilfordii* polyglycoside in combination with glycyrrhizic acid has been demonstrated to reduce liver injury (33). Considering complex diseases, the model of drug discovery has been changed from identifying a single target to identifying multi-targets based on systems biology (34). Notably, herbal remedies

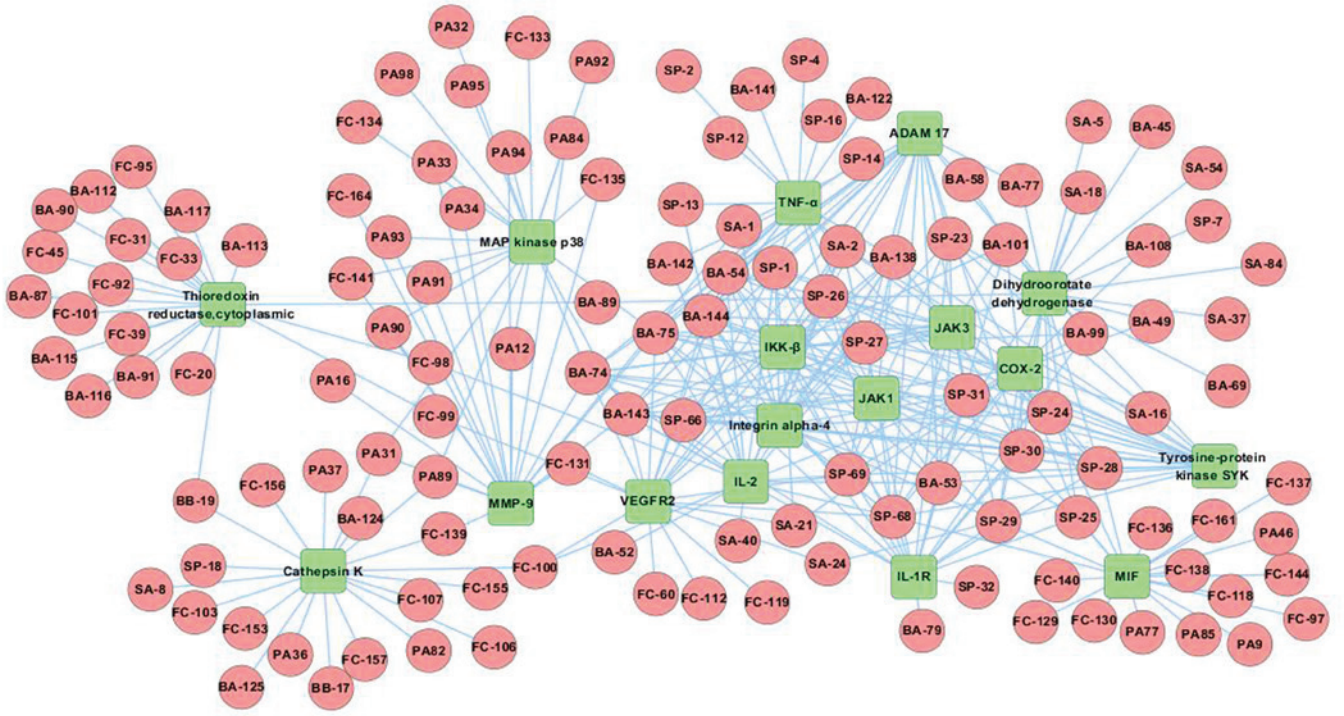

Figure 3. Compound-target network. Potentially active compounds are connected with associated diseases; the circles and rectangles represent the candidate compounds and target proteins, respectively. D, disease; BA, *bile arisaema*; SP, *Semen persicae*; FC, *Flos carthami*; SA, *Sinapis alba*; BB, *Bombyx batryticatus*; PA, *Paeonia alba*.

with the characteristics of multi-component and multi-target compounds are most prevalent and effective in the treatment of chronic illnesses in a number of Asian countries (35); thus, chemical components can be used effectively in Chinese and Western medicines. (36). Thus, it may be useful to study compounds, targets and networks to investigate how Chinese herbal ingredients are effective against RA.

HTTLF is a traditional herbal formula that has been widely used in the Xiamen Hospital of Traditional Chinese Medicine. In the present study, the results demonstrated that the chemical space distributions of compounds from HTTLF were diverse. The data also demonstrated that there is a large overlap between HTTLF compounds and drug/drug-like compounds in the chemical space. According to the QSAR, the compounds with similar chemical space have similar active properties (28). Thus, the compounds from HTTLF may have diverse properties; the majority of compounds possessed drug-like properties, which aids in identifying anti-RA compounds from HTTLF.

Docking results in the current study demonstrated that 127 compounds from HTTLF could interact with 17 targets associated with RA. Among them, 50 compounds had potentially a large number of drug properties, while 77 compounds had the potential to be used in combination therapy. These results demonstrate that HTTLF is a broad-spectrum herbal treatment. To further understand the association between potential compounds and their targets, a CTN was constructed. The network consisted of 144 nodes (127 compounds and 17 targets) and 340 edges. The compounds in the outer CTN displayed fewer interactions with targets than those in the inner CTN. The values of network heterogeneity and network centralization were 1.336 and 0.108, respectively. This indicated that a number of compounds were more central than others (37). For example, isoschaftoside (BA-75) had the largest number of target interactions, whereas β-carotene had one target interaction. Thus, HTTLF exhibited diverse therapeutic effects via interacting with the same or different targets.

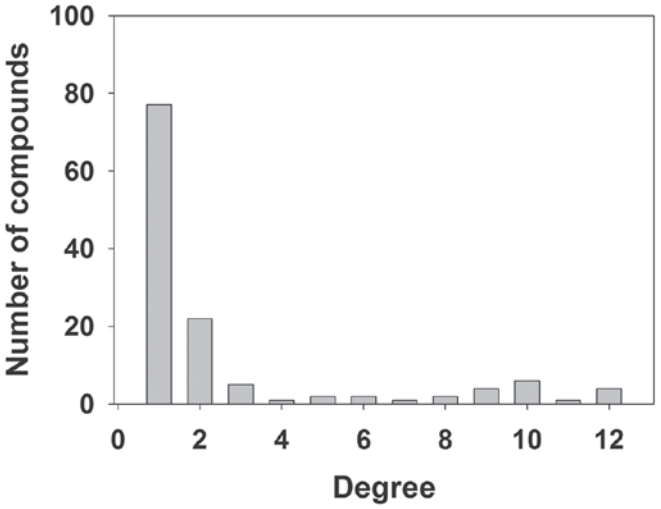

Figure 4. Degree distributions of compound nodes.

Previous studies have demonstrated that particular compounds exhibit biological activities against targets associated with RA (38-41). For example, chlorogenic acid can

Table V. Diseases associated with potential targets.

Index	Disease
D1	Abdominal aortic aneurysm
D2	Acute pain
D3	Adenomatous polyposis
D4	Advanced lung cancer
D5	Allergic rhinitis, unspecified
D6	Allergy, unspecified
D7	Alzheimer's disease
D8	Amebiasis
D9	Arthritis
D10	Asthma
D11	Atherosclerosis
D12	Autoimmune diseases
D13	Bacterial infections
D14	Behcet's disease
D15	Bladder cancer
D16	Brain cancer
D17	Breast cancer
D18	Cancer, unspecific
D19	Cancers
D20	Carcinoma *in situ*, unspecified
D21	Carpal tunnel syndrome
D22	Chronic pain
D23	Colorectal cancer
D24	Congestive heart failure
D25	Coronary artery disease
D26	Diabetes mellitus
D27	Diabetic nephropathy
D28	Dysmenorrhea, unspecified
D29	Endometriosis
D30	Facial Pain
D31	Fibrosing alveolitis
D32	Genitourinary tumors
D33	Gestational hypertension
D34	Glomerulonephritis
D35	Graft-versus-host disease
D36	Guillain-barre syndrome
D37	Heart failure
D38	Hepatocellular carcinoma
D39	Hormone-refractory prostate cancer
D40	Human immunodeficiency virus disease
D41	Hyperimmunoglobulinemia D
D42	Inflammation
D43	Inflammatory bowel disease
D44	Kaposi's sarcoma
D45	Leprosy, unspecified
D46	Lung cancer
D47	Lymphoma, non-hodgkin
D48	Malaria
D49	Malignancies
D50	Malignant mesothelioma
D51	Meningioma
D52	Multiple sclerosis
D53	Myocardial infarction
D54	Neurologic disorders
D55	Non-insulin-dependent diabetes mellitus
D56	Non-small cell lung cancer
D57	Obstructive airway disease
D58	Oropharyngeal squamous cell carcinoma
D59	Osteoarthritis
D60	Osteoporosis
D61	Pain, unspecified
D62	Pancreatic cancer
D63	Parasitic diseases
D64	Pathological angiogenesis
D65	Periodic fever syndrome
D66	Peutz-jeghers syndrome
D67	Pneumocystis infections
D68	Prostate cancer
D69	Psoriasis
D70	Renal cell carcinoma
D71	Restenosis
D72	Rheumatic diseases
D73	Rheumatoid arthritis
D74	Rheumatoid arthritis, unspecified
D75	Sepsis
D76	Septic shock
D77	Sjogren's syndrome
D78	Stroke
D79	Systemic lupus erythematosus
D80	Thrombocytopenia
D81	Tumors
D82	Ulcerative colitis

inhibit the expression of cyclooxygenase-2 and attenuate pro-inflammatory cytokines (including interleukin-1β and tumor necrosis factor-α), which may be beneficial for the prevention and treatment of inflammatory diseases. Quercetin and amygdalin have also been reported to possess anti-inflammation properties (39,41). The combination of these three compounds may have synergistic actions on anti-inflammation. In addition, the CTN demonstrated that the three compounds shared 6 common targets. Thus, the synergistic actions of compounds in HTTLF may be responsible for the therapeutic efficacy of RA.

In order to further verify the diverse functions of HTTLF, a TDN was constructed. The network contained 17 targets and 82 diseases. For example, MMP-9 connected with 15 diseases, including advanced lung cancer, atherosclerosis, brain cancer, osteoarthritis and rheumatoid arthritis. Clinical studies have demonstrated that the expression of MMP-9 can reflect the progression of knee osteoarthritis, atherosclerotic coronary artery disease and RA (42-44). In addition, according to the Medical Subject Headings

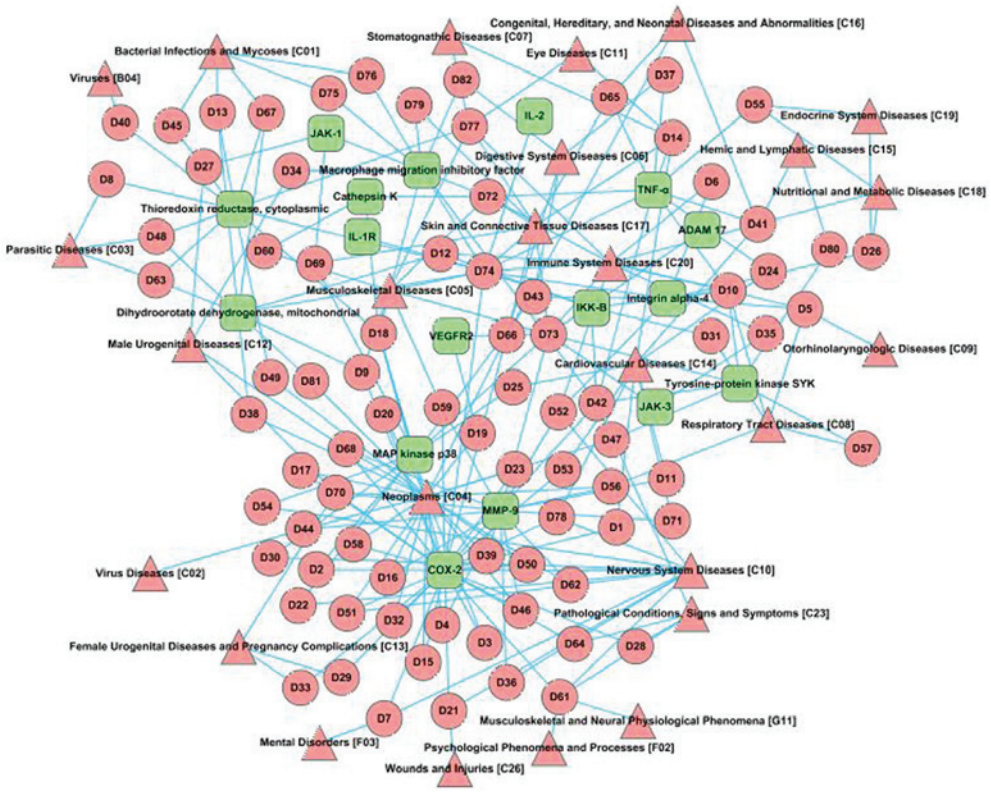

Figure 5. Target-disease network. In the target-disease network, 17 targets (rectangles) were associated with 82 diseases (circles) which belonged to 26 categories (triangles). D, disease.

(http://www.nlm.nih.gov/mesh/MBrowser.html) the 82 diseases were classified into 26 groups, including skin and connective tissue diseases, neoplasms, immune system diseases, cardiovascular diseases and musculoskeletal diseases. For example, osteoarthritis, osteoporosis and RA belong to musculoskeletal diseases; Sjögrkn's syndrome, asthma and allergic rhinitis are immune system diseases. This suggests that compounds from HTTLF have potential therapeutic effects on diverse diseases (45) in addition to RA; for example, amygdalin can be used to treat diseases such as asthma, tumors and diabetes (46). Thus, a TDN provides a visual representation of the association between potential active compounds of HTTLF and diseases through the targets associated with RA.

In conclusion, a novel platform of system pharmacology integrating physicochemical property analysis, active compound prediction, and compound-target and target-disease associate networks was created in the present study, in order to investigate the mechanisms underlying the therapeutic effects of HTTLF on RA. The results demonstrated that: i) Compounds from HTTLF exhibit diverse chemical space and drug-like properties; ii) a total of 127 compounds from HTTLF are regarded as potentially active compounds, interacting with 17 targets associated with RA; iii) compounds from HTTLF have diverse and synergistic actions in the treatment of RA, and exert therapeutic effects for other diseases, such as osteoarthritis, osteoporosis and neoplasms. This is consistent with the hypothesis of the 'same treatment for different diseases' in TCM. The present investigation provides a visual understanding of the chemical and pharmacological basis of TCM; this is beneficial to the discovery of anti-rheumatoid drugs from TCM. Based upon the present study, future research will include the extraction of effective constituents from HTTLF, which will be examined *in vivo* and *in vitro* for the discovery of anti-rheumatoid drugs from TCM.

Acknowledgements

The present study was supported by the National Natural Science Foundation of China (grant no. 81473574) and the Developmental Fund of ChenKeji Integrative Medicine (grant no. CKJ2015010).

References

1. Qin ZJ: The research progress of rheumatoid arthritis in diagnosis and treatment of Chinese and western medicines. Acta Med Sin 24: 224-227,2011. (In Chinese).
2. Fan FY and He XL: Research progress on traditional Chinese medicine for rheumatoid arthritis. Journal of Traditional Chinese Medicine University of Hunan 26: 60-61, 2006 (In Chinese).
3. Tong G, Zhang X, Tong W and Liu Y: Association between polymorphism in STAT4 gene and risk of rheumatoid arthritis: A meta-analysis. Hum Immunol 74: 586-592, 2013.
4. Xu J, Wang Q and Liu SY: Drug treatment of rheumatoid arthritis. Chin Community Doctors 17: 22-23, 2002. (In Chinese).
5. Niu LJ: Pharmacotherapy of rheumatoid arthritis. Pain Clin J 2: 106-109, 2006 (In Chinese).
6. Li QX: Research progress on immune mechanisms of traditional Chinese medicine for the treatment of rheumatoid arthritis. Research of Traditional Chinese Medicine 18: 53-55, 2002 (In Chinese).
7. Huang MC, Pai FT, Lin CC, Chang CM, Chang HH, Lee YC, Sun MF and Yen HR: Characteristics of traditional Chinese medicine use in patients with rheumatoid arthritis in Taiwan: A nationwide population-based study. J Ethnopharmacol 176 :9-16, 2015.

8. Wang JM, Tao QW, Zhang YZ, Xu Y and Yan XP: Treating rheumatoid arthritis patients of Shen deficiency and cold invading syndrome by bushen quhan zhiwang decoction combined methotrexate: An evaluation of clinical efficacy and safety. Zhongguo Zhong Xi Yi Jie He Za Zhi 33: 614-618, 2013.(In Chinese).
9. Chen ZH: Clinical observation of Yishen Zhuanggu Tongbi Decoction in the Treatment of rheumatoid arthritis. Journal of Guangzhou University of Traditional Chinese Medicine 22: 19-22, 2005 (In Chinese).
10. Zhang YY, Chen JC, Qiu MS and Xu ZX: Clinical Observation of Huatan Tongluo Fang in the treatment of rheumatoid arthritis with phlegm-obstruction and blood-stasis Type. Traditional Chinese Medicine Journal 11: 49-52, 2012 (In Chinese).
11. Qiu MS, Chen JC, Zhang YY, Xu ZX, Jiang YF and Peng YF: Effect of Huatan Tongluo Fang on collagen-induced arthritis in rats. Journal of Beijing University of Traditional Chinese Medicine 35: 121-124, 2012 (In Chinese).
12. Zheng CS, Ye HZ, Xu XJ and Liu XX: Computational pharmacology study of tougu xiaotong granule in preventing and treating knee osteoarthritis. Chin J Integr Med 15: 371-376, 2009.
13. Zheng CS, Xu XJ, Ye HZ, Wu GW, Li XH, Huang SP and Liu XX: Computational approaches for exploring the potential synergy and polypharmacology of Duhuo Jisheng Decoction in the therapy of osteoarthritis. Mol Med Rep 7: 1812-1818, 2013.
14. Shi SH, Cai YP, Cai XJ, Zheng XY, Cao DS, Ye FQ and Xiang Z: A network pharmacology approach to understanding the mechanisms of action of traditional medicine: Bushenhuoxue formula for treatment of chronic kidney disease. PLoS One 9: e89123, 2014.
15. Qiao X, Hou T, Zhang W, Guo S and Xu X: A 3D structure database of components from Chinese traditional medicinal herbs. J Chem Inf Comput Sci 42: 481-489, 2002.
16. Zhou JJ, Xie GR and Yang JX (eds): Handbook of the Chemical Constituents in Chinese Herb Original Plants. Chemical Industry Press, Beijing, pp1165-pp1211, 2004 (In Chinese).
17. Wang ZQ: Research progress of chemical composition and pharmacological effects of Rhizoma arisaematis. Drug Evaluation Research 32: 144-149, 2009 (In Chinese).
18. Zheng CS, Xu XJ, Ye HZ, Wu GW, Li XH, Xu HF and Liu XX: Network pharmacology-based prediction of the multi-target capabilities of the compounds in Taohong Siwu decoction and their application in osteoarthritis. Exp Ther Med 6: 125-132, 2013.
19. Cheng SM, Wang HY, Li GY, Zhu Y, Zhao WB and Wang JH: Research progress of Bombyx batryticatus. Journal of Nongken Medicine 34: 443-448, 2012 (In Chinese).
20. Sheridan RP and Shpungin J: Calculating similarities between biological activities in the MDL Drug Data Report database. J Chem Inf Comput Sci 44: 727-740, 2004.
21. Hassan M, Bielawski JP, Hempel JC and Waldman M: Optimization and visualization of molecular diversity of combinatorial libraries. Mol Divers 2: 64-74, 1996.
22. Lipinski CA, Lombardo F, Dominy BW and Feeney PJ: Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. Adv Drug Deliv Rev 46: 3-26, 2001.
23. Zhu F, Shi Z, Qin C, Tao L, Liu X, Xu F, Zhang L, Song Y, Liu X, Zhang J, *et al*: Therapeutic target database update 2012: A resource for facilitating target-oriented drug discovery. Nucleic Acids Res 40: D1128-D1136, 2012.
24. Hashimoto N, Iwasaki T, Kitano M, Ogata A and Hamano T: Levels of vascular endothelial growth factor and hepatocyte growth factor in sera of patients with rheumatic diseases. Mod Rheumatol 3: 129-134, 2003.
25. Rose PW, Prlić A, Bi C, Bluhm WF, Christie CH, Dutta S, Green RK, Goodsell DS, Westbrook JD, Woo J, *et al*: The RCSB Protein Data Bank: Views of structural biology for basic and applied research and education. Nucleic Acids Res 43 (D1): D345-D356, 2015.
26. Montes M, Braud E, Miteva MA, Goddard ML, Mondésert O, Kolb S, Brun MP, Ducommun B, Garbay C and Villoutreix BO: Receptor-based virtual ligand screening for the identification of novel CDC25 phosphatase inhibitors. J Chem Inf Model 48: 157-165, 2008.
27. Smoot ME, Ono K, Ruscheinski J, Wang PL and Ideker T: Cytoscape 2.8: New features for data integration and network visualization. Bioinformatics 27: 431-432, 2011.
28. Dobson CM: Chemical space and biology. Nature 432: 824-828, 2004.
29. Tanaka Y: Current concepts in the management of rheumatoid arthritis. Korean J Intern Med 31: 210-218, 2016.
30. Wang NN, Wang AW and Hao QX: New progress of study on traditional Chinese medicine for the treatment of rheumatoid arthritis. Modern Journal of Integrated Chinese Traditional and Western Medicine 20: 2334-2336, 2011 (In Chinese).
31. Zheng CE: The experience of research of integrated traditional Chinese and Western medicine for the treatment of rheumatoid arthritis. Integrated Traditional Chinese and Western Medicine 4: 255-256, 2012 (In Chinese).
32. Wu YJ, Lao ZY and Zhang ZL: Clinical observation on small doses *Tripterygium wilfordii* polyglycoside combined with methotrexate in treating rheumatoid arthritis. Zhongguo Zhong Xi Yi Jie He Za Zhi 21: 895-896, 2001. (In Chinese).
33. Li YS,Yang JH,Fan SY and Zhang YY:The experimental study of *Tripterygium wilfordii* polyglycoside combination therapy with glycyrrhizic acid for rheumatoid arthritis on efficacy enhancing and toxicity reducing. Chin J Tradit Med Sci Technol 15:293, 2008. (In Chinese).
34. Kitano H: A robustness-based approach to systems-oriented drug design. Nat Rev Drug Discov 6: 202-210, 2007.
35. Chen M, Feng W, Cao H, Zou L, Chen C, Baatrup A, Nielsen AB, Li H, Kassem M, Zou X and Bünger C: A traditional Chinese medicine formula extracts stimulate proliferation and inhibit mineralization of human mesenchymal stem cells in vitro. J Ethnopharmacol 125: 75-82, 2009.
36. Li XD, Huang Q and Xu XJ: Target-Li gand space distribution of the compound sets from Xuefu Zhuyu Decoction, a traditional chinese medicinal recipe. Acta Physico-Chimica Sinica 24: 547-550, 2008 (In Chinese).
37. Ma JL, An LL and Xing X: Topology analysis on urban public traffic networks. Journal of Shanxi Teachers University 23: 51-55, 2009 (In Chinese).
38. Yang B, Qiu Y, Wang LP and Zhang XL: Studies on the anti-inflammatory molecular mechanism of chlorogenic acid extracted from *Lonicera confusa* DC in vitro. Chinese Pharmacological Bulletin 25: 542-545, 2009 (In Chinese).
39. Huang JQ, Sun WJ, Wang SW and Zhu MZ: Studies on the anti-inflammatory and antioxidant activity of quercetin in rats with gouty arthritis. China Journal of Experiment Traditional Medical Formulae 18: 169-173, 2012 (In Chinese).
40. Wang MY, Wei YF and Li XB: Chemical constituents of anti-rheumatism fraction from Urtica fissa. Chinese Traditional and Herbal Drugs 37: 1300-1303, 2006 (In Chinese).
41. Lv JZ and Deng JG: Research progress in pharmacological effects of amygdalin. Drugs & Clinic 27: 530-535, 2012 (In Chinese).
42. He ZK and Shen JW: The value of MMP-2, MMP-3, MMP-9 and TIMP-1 levels in the evaluation of knee joint osteoarthritis. Chongqing Medical Journal 42: 3872-3874, 2013 (In Chinese).
43. Wu ZB, Lu N, Wang YH, Shi ZG, Fan CM, Jia J, Liu L, Leng N and Zhu P: Association of serum MMP-2 and MMP-9 and progression of joint damage in early rheumatoid arthritis. Chinese Journal of Immunology 22: 260-262, 2006 (In Chinese).
44. Gao F, Chen LL, Yan SR, Guo SR, Zhao LH, Yang P, Zhang CW and Ji XB: Correlation between serum levels of MMP-1, MMP-9, TIMP-1, IL-6 and the severity of atherosclerotic coronary artery disease. Journal of Clinical Research 30: 2384-2386, 2013 (In Chinese).
45. Royall J and Lyon B: Sea-change or change challenge? Health information access in developing countries: The U.S. National Library of Medicine experience. Afr Health Sci 11:457-463,2011.
46. Zhou C, Qian L, Ma H, Yu X, Zhang Y, Qu W, Zhang X and Xia W: Enhancement of amygdalin activated with β-D-glucosidase on HepG2 cells proliferation and apoptosis. Carbohydr Polym 90: 516-523, 2012.