2015

Saussurea involucrata: A review of the botany, phytochemistry and ethnopharmacology of a rare traditional herbal medicine

Wai-I Chik
Hong Kong Baptist University

Lin Zhu
Hong Kong Baptist University

Lan-Lan Fan
Guangxi Botanical Garden of Medicinal Plant

Tao Yi
Hong Kong Baptist University, yitao@hkbu.edu.hk

Guo-Yuan Zhu
Macau University of Science and Technology

See next page for additional authors

Authors
Wai-I Chik, Lin Zhu, Lan-Lan Fan, Tao Yi, Guo-Yuan Zhu, Xiao-Jun Gou, Yi-Na Tang, Jun Xu, Wing-Ping Yeung, Zhong-Zhen Zhao, and Zhi-Ling Yu

Saussurea involucrata: A review of the botany, phytochemistry and ethnopharmacology of a rare traditional herbal medicine

Wai-I Chik [a, 1], Lin Zhu [a, 1], Lan-Lan Fan [b, 1], Tao Yi [a *], Guo-Yuan Zhu [c], Xiao-Jun Gou [d], Yi-Na Tang [a], Jun Xu [a], Wing-Ping Yeung [a], Zhong-Zhen Zhao [a], Zhi-Ling Yu [a], Hu-Biao Chen [a, *]

Affiliation

[a] School of Chinese Medicine, Hong Kong Baptist University, Hong Kong Special Administrative Region, China

[b] Guangxi Botanical Garden of Medicinal Plant, Nanning, Guangxi 530023, China

[c] The State Key Laboratory of Quality Research in Chinese Medicine, Macau University of Science and Technology, Macau, China

[d] Sichuan Industrial Institute of Antibiotics, Chengdu University, Chengdu, 610051, China

[*] Corresponding author at: School of Chinese Medicine, Hong Kong Baptist University, Hong Kong Special Administrative Region, P. R. China. Tel.: +852 3411 2081, +852 3411 2060; fax: +852 3411 5571, +852 3411 2461.

E-mail address: yitao@hkbu.edu.hk (T. Yi); hbchen@hkbu.edu.hk (H. B. Chen).

[1] These authors contributed equally to this work.

ABSTRACT

Ethnopharmacological relevance: *Saussurea involucrata* Matsum. & Koidz. is an endangered species of the Asteraceae family, growing in the high mountains of central Asia. It has been, and is, widely used in traditional Uyghur, Mongolian and Kazakhstan medicine as well as in Traditional Chinese Medicine as Tianshan Snow Lotus (Chinese: 天山雪蓮). In traditional medical theory, *S. involucrata* can promote blood circulation, thereby alleviating all symptoms associated with poor circulation. It also reputedly eliminates cold and dampness from the body, diminishes inflammation, invigorates, and strengthens *Yin and Yang*. It has long been used to treat rheumatoid arthritis, cough with cold, stomachache, dysmenorrhea, and altitude sickness in Uyghur and Chinese medicine.

Aim of the review: To comprehensively summarize the miscellaneous research that has been done regarding the botany, ethnopharmacology, phytochemistry, biological activity, toxicology of *S. involucrata.*

Method: An extensive review of the literature was carried out. Apart from different electronic databases including SciFinder, Chinese National Knowledge Infrastructure (CNKI), ScienceDirect that were sourced for information, abstracts, full-text articles and books written in English and Chinese, including those traditional records tracing back to the *Qing Dynasty*. Pharmacopoeia of China and other local herbal records in Uighur, Mongolian and Kazakhstan ethnomedicines were investigated and compared for pertinent information.

Results: The phytochemistry of *S. involucrata* has been comprehensively investigated. More than 70 compounds have been isolated and identified; they include phenylpropanoids, flavonoids, coumarins, lignans, sesquiterpenes, steroids, ceramides, polysaccharides. Scientific studies on the biological activity of *S. involucrata* are equally numerous. The herb has been shown to have anti-neoplastic, anti-inflammatory, analgesic, anti-oxidative, anti-fatigue, anti-aging, anti-hypoxic, neuroprotective and immunomodulating effects. Many have shown correlations to the traditional clinical applications in Traditional Chinese Medicine and medicines. The possible mechanisms of *S. involucrata* in treating various cancers are revealed in the article, these include inhibition of cancer cells by affecting their growth, adhesion, migration, aggregation and invasion, inhibition of epidermal growth factor receptor signaling in cancer cells, hindrance of cancer cell proliferation, causing cytotoxicity to cancer cells and promoting expression of tumor suppressor genes. Dosage efficacy is found to be generally concentration- and time-dependent. However, studies on the correlation between particular chemical constituents and specific bioactivities are limited.

Conclusion: In this review, we have documented the existing traditional uses of *Saussurea involucrata* and summarized recent research into the phytochemistry and pharmacology of *Saussurea involucrata*. Many of the traditional uses have been validated by phytochemical and modern pharmacological studies but there are still some areas where the current knowledge could be improved. Although studies have confirmed that *Saussurea involucrata* has a broad range of bioactivities, further in-depth studies on the exact bioactive molecules and the mechanism of action are expected. Whether we should use this herb independently or in combination deserves to be clarified. The exact quality control as well as the toxicology studies is necessary to guarantee the stability and safety of the clinic use. The sustainable use of this endangered resource was also addressed. In conclusion, this review was anticipated to highlight the importance of *Saussurea involucrata* and provides some directions for the future development of this plant.

Keywords:
Saussurea involucrata; Traditional Chinese Medicine; Uyghur local medicine; Phytochemistry; Ethnopharmacology; Rheumatoid arthritis

Abbreviations:
DSE, dark-septate endophytic; SEM, scanning electron microscope; RA, rheumatoid arthritis; NO, nitric oxide; PGE_2, prostaglandin E_2; ROS, reactive oxygen species; DPPH, 2, 2-diphenyl-1-picrylhydrazyl; ABTS, 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid); HIF-1, hypoxia-inducible factor-1; ATP, adenosine triphosphate; LAC, laccase; LD, lactate dehydrogenase; SOD, superoxide dismutase; MDA, malondialdehyde; GPx, glutathione peroxidase; PBMC, peripheral blood mononuclear cell.

Contents

1. Introduction

In central Asia, *Saussurea involucrata* Matsum. & Koidz (**Fig. 1.**) has long been used under the herbal names "Tianshan Snow Lotus", “Xinjiang Xuelian”, “Xuelian Hua” and “Xuehe Hua” (Flora of China Committee, 1999). *S. involucrata* has been an important medicinal herb in various ethnomedical systems which are namely Traditional Chinese Medicine, Uyghur medicine, Mongolian medicine and Kazakhstan medicine, among which similarities in pattern of usage has been manifested. These include treatment of rheumatoid arthritis and regulation of menstrual cycle. It is recorded in Pharmacopoeia of Peoples’ Republic of China since 2005 and earlier in local herbal records of the above mentioned ethnomedicines (Chinese Pharmacopoeia Commission, 2010; The National Insitute for Food and Drug Control of China, 1984).

Fig. 1. Photos of *Saussurea involucrata* plant and its medicinal material.

According to the Guangdong Provincial Traditional Chinese Medicine Hospital, which is the top Traditional Chinese Medicine hospital in China, *S. involucrata* has a broad-spectrum of clinical applications, including anti-inflammatory and analgesic, anti-oxidative, anti-hypoxia, anti-fatigue, anti-aging and hormonal-related gynecological disorders, infertility as well as immunomodulation.

These are closely correlated with those traditional uses. In recent years, the effectiveness in anti-cancer therapy has further put *S. involucrata* under the spotlight.

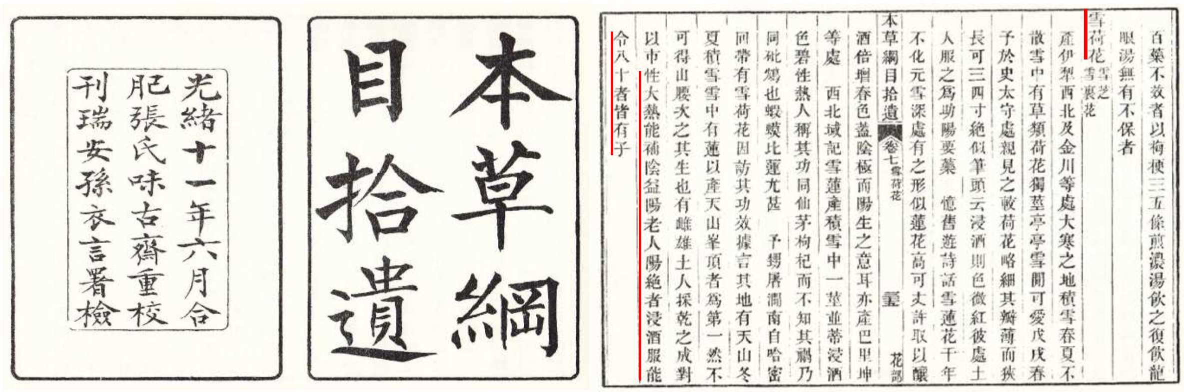
Along with its growing reputation, chemical constituents of *Saussurea involucrata* have been extensively studied. Phenylpropanoids, flavonoids, coumarins, lignans, sesquiterpenes, steroids, ceramides and polysaccharides as main compositions were isolated and identified. Among them, rutin and chlorogenic acid have been proven with pharmacological efficacy and active compounds in abundance of *S. involucrata* (Yi et al., 2009b) and they are the chemical markers for its quality control in the current version of Chinese Pharmacopoeia (Chinese Pharmacopoeia Commission, 2010).

In this review, advances in ethnopharmacology in different ethnomedical systems, phytochemistry, biological and pharmacological activities, toxicology and clinical application of *S. involucrata* are revealed.

2. Ethnopharmacology

The dried aerial parts of *Saussurea involucrata* have long been used as an herbal medicine in different parts of China and Kazakhstan. The use of *Saussurea involucrata* in local medicines in Central Asia has a long history (**Table 1**). However, the earliest scientific record can only be traced back to the Qing Dynasty (from 1636 A.D. to 1912 A.D.). The earliest literature on *Saussurea involucrata* was in Bencao Gangmu Shiyi, in English 'Supplement to Compendium of Materia Medica' (Zhao, 1963), in which *Saussurea involucrata* can be used to nourish *Yin and Yang*, which is considered the origin of life and materials to maintain balance in body in Traditional Chinese Medicine theory, treat diseases related to internal coldness (**Fig. 2**). It can also promote fertility in aged people and enhance recovery from measles (Xie, 1968; Zhao, 1963). Impotence in men can also be improved by using the decoction of 6 g of Herba Saussureae Involucratae together with 3 g of Radix Angelicae Sinensis and Fructus Lycii each (Committee for National Revolution of

Xinjiang Uyghur Autonomous Region Health Bureau, 1976).



光緒十一年六月合
肥張氏味古齋重校
刊瑞安孫衣言署檢

本草綱目拾遺

百藥不效者以梅使三五條煎濃湯飲之復飲龍
眼湯無有不保者
雪荷花 雪芝 雪裏花
產伊犁西北及金川等處大寒之地積雪春夏不
散雪中有草類荷花獨莖亭亭雪間可愛戊戌春
予於史太守處親見之較荷花略細其瓣薄而狹
長可三四寸絶似筆頭云浸酒則色微紅彼處土
人服之爲助陽要藥　憶舊遊詩話雪蓮花千年
不化元雪深處有之形似蓮花高可丈許取以釀
本草綱目拾遺 卷七 雪荷花 花部
酒倍增春色蓋陰極而陽生之意耳亦產巴里坤
等處　西北域記雪蓮產積雪中一莖並蒂浸酒
色碧性熱人稱其功同仙茅枸杞而不知其禍乃
同砒鴆也蝦蟆比蓮尤甚　予甥居滇南自哈密
回帶有雪荷花因詢其功效據言其地有天山冬
夏積雪雪中有蓮以產天山峯頂者爲第一然不
可得山腰次之其生也有雌雄土人採乾之成對
以市性大熱能補陰益陽老人陽絶者浸酒服能
令八十者皆有子

Fig. 2. The earliest literature on *Saussurea involucrata* (in Bencao Gangmu Shiyi book)

The extensive use of *Saussurea involucrata* has been demonstrated in different ethnomedical systems including Uyghur medicine, Mongolian medicine, Kazakhstan medicine and Traditional Chinese Medicine (**Table 1**). Amazingly, similarities in clinical applications among different local medicines were observed in treatment of gynecological disorders, reliving respiratory symptoms and pain-killing. It was worth to point out that unlike other herbs in Traditional Chinese Medicines, in traditional herbal records, *Saussurea involucrata* is usually used independently instead of in herbal formulas having prescription theories as “monarch, minister, assistant and guide’ (Chinese Medicinal Materials Corporation, 1994).

Table 1 The traditional uses of *Saussurea involucrata* in different ethnomedical systems.

	Traditional Chinese Medicine	Uyghur medicine	Mongolian medicine	Kazakhstan medicine	References
Herbal name	"Tianshan Xuelian" (Chinese: 天山雪蓮)	"Tage Leylishi" (Uyghur: 塔古來力斯)	"Banzhi Dawa" (Mongolian: 幫孜達娃)	"Huojia Quepu" (Kazakhstan: 霍加却僕)	(Chinese Pharmacopoeia Commission, 2010; Jia et al., 2005)
Part(s) of herb used	Aerial parts	Whole part	Aerial parts	Not mentioned	
Dosage	3~6 g				
Indications on pregnancy	Prohibited	Prohibited	Not mentioned	Not mentioned	

Independent Use

Drug Form	Clinical application	Traditional Chinese Medicine	Uyghur medicine	Mongolian medicine	Kazakhstan medicine	References
Decoction/ Tincture	Rheumatoid arthritis	√	√			(Chinese Pharmacopoeia Commission, 2010; Committee for National Revolution of Xinjiang Uyghur Autonomous Region Health Bureau, 1976; Jia et al., 2005; Liu and Shawuti, 1985; Commission of Chinese Materia Medica, 1999; Xie Z.W., 1975; National Institutes for Food and Drug Control, 1984)
	Menstrual pain	√	√	√		(Chinese Medicinal Materials Corporation, 1994; Chinese Pharmacopoeia Commission, 2010; Jia et al., 2005; Liu and Shawuti, 1985; Commission of Chinese Materia Medica, 1999; National Institutes for Food and Drug Control, 1984)
	Irregular menstruation cycle	√	√	√		(Chinese Medicinal Materials Corporation, 1994; Chinese Pharmacopoeia Commission, 2010; Jia et al., 2005; Liu and Shawuti, 1985; Commission of Chinese Materia Medica, 1999; Xie Z.W., 1975; National Institutes for Food and Drug Control, 1984)
	Infertility	√				(Chinese Pharmacopoeia Commission, 2010; Xie Z.W., 1975; Zhao, 1963)
	Labour-inducing agent				√	(Jia et al., 2005; Xie Z.W., 1975)
	Cough	√	√		√	(Jia et al., 2005; Commission of Chinese Materia Medica, 1999; Xie Z.W., 1975)

Decoction	Other pain		√	√	√	(Jia et al., 2005; Liu and Shawuti, 1985; National Institutes for Food and Drug Control, 1984)
	Tuberculosis			√		(Jia et al., 2005)
	Measles	√	√			(Chen, 1963; Commission of Chinese Materia Medica, 1999; Zhao, 1963)
	Strain			√		(Jia et al., 2005)
Snow lotus injection (*i.m.*)	Rheumatoid arthritis; Inflammation	√	√			(Commission of Chinese Materia Medica, 2005)

Classic Formula

Formula Name	**Ingredients**	**Clinical application**	**Traditional Chinese Medicine**	**Uyghur medicine**	**Mongolian medicine**	**Kazakhstan medicine**	**References**
Xuelian Tincture	Herba Saussureae Involucratae, Radix Angelicae Sinensis, Fructus Lycii	Menstrual pain, irregular menstruation cycle, infertility	√	√			(Committee for National Revolution of Xinjiang Uyghur Autonomous Region Health Bureau, 1976; Commission of Chinese Materia Medica, 1999)
Xuelian Honghua Tincture	Herba Saussureae Involucratae, Flos Carthami, Fructus Mori, Fructus Lycii	Immunodeficiency, poor blood circulation		√			(Commission of Chinese Materia Medica, 2005)
Xuelian Lingzhi Tincture	Herba Saussureae Involucratae, Ganoderma	Infertility, immunodeficiency		√		√	(Commission of Chinese Materia Medica, 2005)

Xuelian Patches (Protected formula)	Herba Saussureae Involucratae	Anti-neoplastic agent	√	√			(Commission of Chinese Materia Medica, 2005)
Other commonly used formula							
Formula Name	**Ingredients**	**Clinical application**	**Traditional Chinese Medicine**	**Uyghur medicine**	**Mongolian medicine**	**Kazakhstan medicine**	**References**
Xuelian Tincture	Herba Saussureae Involucratae , Flos Carthami, Radix Gentianae Macrophyllae, Radix Angelicae Pubescentis, Fructus Lycii	Arthritis, pain	√	√			(Orally transmitted)
Xuelian Fengshiling Mixture	Herba Saussureae Involucratae, Processed Radix Aconiti, Radix Angelicae Pubescentis	Rheumatoid arthritis, arthritis-related pain, inflammation	√				(Orally transmitted)
Xuelian Guiqi Decoction	Herba Saussureae Involucratae, Radix Angelicae Sinensis, Radix Astragali	Forgetfulness, insomnia, dizziness, exhaustion	√				(Orally transmitted)
Xuelian Chongcao Mixture	Herba Saussureae Involucratae, Cordyceps	Polyuria, exhaustion	√				(Orally transmitted)

3. Botany

3.1 Nomenclature

According to “The Plant List”, *Saussurea involucrata* Matsum. & Koidz is the only accepted name for the herb, with a synonym “*Saussurea involucrata* (Kar. & Kir.) Sch. Bip.” (The Plant List, 2013). However, being the more widely accepted name in China, “*Saussurea involucrata* Kar. & Kir.” has been used as the official name instead for the herb in Pharmacopoeia of Peoples’ Republic of China and Flora of China (Chinese Pharmacopoeia Commission, 2010; Flora of China Editorial Committee, 1999).

A recent review has indicated the importance of accurate scientific nomenclature for plants so as to minimize ambiguity of species (Rivera et al., 2014) and therefore, the confusion concerning *Saussurea involucrata* mentioned above is expected to be resolved.

3.2 Plant occurrence

The habitats of *Saussurea involucrata* include mountain slopes, mountain valleys, meadows and rock fissures at elevations of 2,400 - 4,100 m. Due to the incredibly high altitudes at which it grows; *Saussurea involucrata* is named “Tianshan Snow Lotus” which means the Snow Lotus that grows on sky-high mountains. The species is mainly distributed in Xinjiang of China, but it also occurs in Kazakhstan, Kyrgyzstan and Mongolia (Chen et al., 1999).

Unfortunately, due to over-exploitation, the wild population of *Saussurea involucrata* is dwindling rapidly (Kang et al., 2010). According to the People's Daily, *Saussurea involucrata* has decreased from being found over 50,000,000 acres in the 1960’s and 1970’s to a few hundred acres at present. At this rate, in less than 10 years, this rare species could become extinct. It has thus been listed as a second grade national protected wild plant in China (Fu and Jin, 1991).

3.3 Botanical description

Saussurea involucrata is a perennial herb 15-50 cm tall. It has an unbranched, stout caudex which is densely covered with fibrous remains of petioles. Its stem is solitary with its rosette and stem leaves petiolated. The leaf blades are narrowly ovate, elliptic, or obovate, with both surfaces being green and glandular hairy. The uppermost stem leaves are sessile, ovate to elliptic, being membranous with both surfaces pale yellow; and stellately surround the synflorescence. The synflorescence is hemispheric to broadly campanulate with 10-20 capitula. Its phyllaries are in 3 or 4 rows, dark or light brown with dark margins and sparsely pubescent. The achenes are straw-colored with blackish spots, cylindrical, and each has a dirty-white pappus. Plants flower from July to August and bear fruit from August to October (Flora of China Editorial Committee, 1999).

3.4 Conservation

Saussurea involucrata is a valuable medicinal herb and it has also been an important source of a wide range of bioactive compounds; syringin, chlorogenic acid, and 1, 5-dicaffeoylquinic acid are the chemicals most commonly extracted from it (Chen et al., 2014b). Wild populations of *Saussurea involucrata* are dwindling rapidly. To reduce the demand for the wild *Saussurea involucrata*, researches have been done to increase yield of chemicals by modified cultivation method, developing tissue culture systems for its rapid propagation and finding substitutes.

According to a study, the increasing duration of growth in the cultivation of *Saussurea involucrata* can increase the amount of some of its bioactive compounds (Y. R. Chen et al., 2013). Besides, a dark-septate endophytic (DSE) fungus EF-37 isolated from the roots of *Saussurea involucrata* has also demonstrated a significant positive effect on plant growth and its rutin content (Wu and Guo, 2008; Wu et al., 2010), though with withheld mechanism. Optimization in conditions of micropropagation systems for maximization of chemicals having desirable biocativities have been investigated, such as the reduction of atmospheric pressure which has resulted in variation of morphogenic potential and anti-oxidative enzymatic activities (Guo et al., 2011, 2007). According to one study, hairy root culture system of *Saussurea involucrata* through agrobacterium-mediated transformation was established and the amount of syringin and hispidulin, two of the active components, was found to be even higher than in the wild species (Fu et al., 2006; Qiu et al., 2010). These suggest the feasibility of using tissue culture of *Saussurea involucrata* to replace its wild plant source.

Closely-related plant species often share similar secondary metabolites and bioactivities (Cock et al., 2010). With reference to a recent review on the genus, the *Saussurea* shows resemblance in phytochemical constituents as well as bioactivities. Dehydrocostus lactone (Li and Jia, 1989; Madhavi et al., 2012), 3a,8a-dihydroxy-11bH-11,13-dihydrodehydrocostus lactone (Li and Jia, 1989), 35,36, 3a,8a-dihydroxy-11bH-11,13-dihydrodehydrocostus lactone 8-*O*-β-D-glucopyranoside (Fan et al., 2006; Li and Jia, 1989), 8a-hydroxy-11bH-11,13-dihydrodehydrocostus lactone (Fan et al., 2006; Li and Jia, 1989) are phytochemicals present in *Saussurea involucrata*, but are also found in other close species including *S. lappa*, or other easily confused species of *Saussurea involucrata* like *S. medusa* or *S. laniceps* (Wang et al., 2010). Besides, *S. ussuriensis*, *S. petrovii*, *S. costus* and *S. medusa* have similar pharmacological function in anti-rheumatic arthritis (Wang et al., 2010). These can serve as evidence for the possible substitution of herbal resources by their closely-related species. Cock has pointed out, which would provide insight into new direction of research as in principal active compound identification, common phytochemicals are good targets for bioactivity testing when one or more species within a genus are known to possess similar therapeutic properties (Cock et al., 2010).

4. Quality control

4.1 Species authentication

Snow Lotus prepared by herbs of different species or different places of origin contains different amount of chemical constituents and thus give rise to different medical value. Therefore, quality control is crucial, for both drug efficacy and safety. There are confusions in the species of herbs contributing to the Chinese Materia Medica “Snow Lotus”. Three representative easily-confused species include *Saussurea involucrata*, *Saussurea laniceps*. and *Saussurea medusa* (Chen et al., 2014a). According to the Pharmacopoeia of China, *Saussurea involucrata* is the only species being accepted (Chinese Pharmacopoeia Commission, 2010). Therefore, traditional and contemporary authentication methods are required to ensure the use of the official species. As demonstrated by a study, the confused species can be differentiated using macroscopic identification of the crude drug, microscopic identification of the powered crude drug and a combination of microscopic identification methods of its pollen grains including ordinary light microscopy, polarized light microscopy and SEM (Chen et al., 2014a).

4.2 Limits of indicator compounds

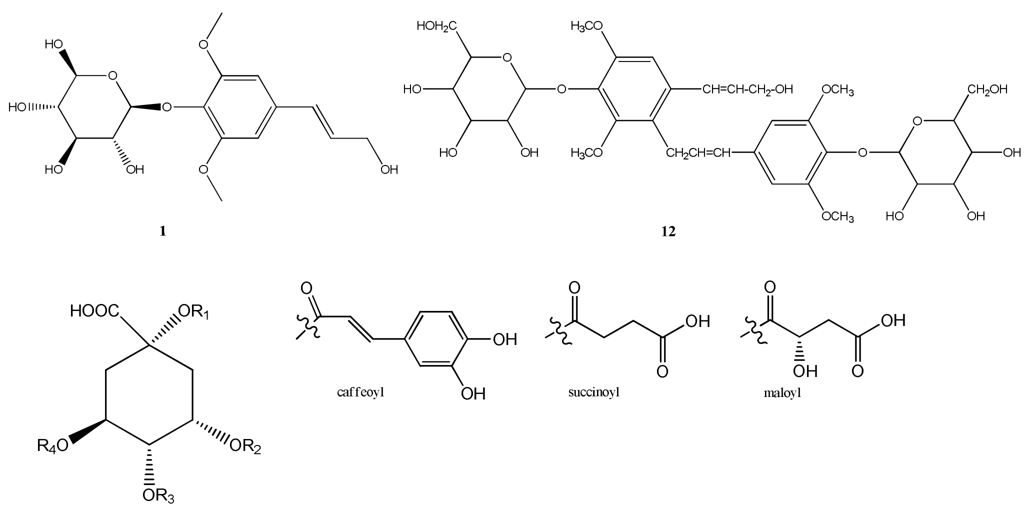
In the view of its chemical content, chlorogenic acid (not less than 0.15% of dried sample) and rutin (not less than 0.15% of dried sample), being the major chemical constituents of *Saussurea involucrata*, are used as the indicator compound to characterize the quality of this herb (Chinese Pharmacopoeia Commission, 2010). The abundance of chemical constituents differs in herbs obtained from different origins. It is believed that the quality of Snow Lotus sourced from Tianshan of Xinjiang in China (Yi et al., 2009b) is the best of all.

5. Phytochemistry

More than 70 compounds have so far been isolated and identified from *Saussurea involucrata*. As aerial parts of *Saussurea involucrata* have been used as in Traditional Chinese Medicine, other folk medicine systems and remain the only medicinal part till now, chemical constituent analysis mainly focused on its above-ground portion. HPLC-MS has been the most common analytical means for qualitative analysis of chemicals present in *Saussurea involucrata*; a diversity of compounds were identified including phenylpropanoids, flavonoids, coumarins, lignans, sesquiterpenes, steroids, ceramides and polysaccharides. In quantitative analysis of the chemicals, which is not as comprehensive as qualitative analysis, HPLC was mainly used. The contents of chlorogenic acid and rutin, which have been set for quality control for *Saussurea involucrata* in the Chinese Pharmacopoeia 2010 edition, ranged from 1.19 to 14.43 mg/g and 0.14 to 16.02 mg/g respectively. Total phenolic content was found to be ranging from 23.58 to 41.56 mg/g while the content for total flavonoid is 9.08 and 15.84 mg/g. Other bioactive compounds were qualitatively determined as well, including syringin (0.20-0.58 mg/g), arctiin (0.05-1.62 mg/g), apigenin (0.005-0.015 mg/g) and hispidulin (0.05-0.16 mg/g) (R. Chen et al., 2013; Li and Zhong, 2013; Qiu et al., 2010; Xu et al., 2009; Yi et al., 2009b).

5.1 Phenylpropanoids

Syringin (**1**), 3-caffeoylquinic acid (**2**) 5-caffeoylquinic acid (**3**), 4-caffeoylquinic acid (**4**), 1,3-dicaffeoylquinic acid (**5**), 1,4-dicaffeoylquinic acid (**6**), 1,5-dicaffeoylquinic acid (**7**), 4,5-dicaffeoylquinic acid (**8**), 1,5-dicaffeoyl-3-succinoylquinic acid (**9**), 1,5-dicaffeoyl-4-succinoylquinic acid (**10**), 1,5-dicaffeoyl-3,4-disuccinoylquinic acid (**11**) have been found in *Saussurea involucrata* (R. Chen et al., 2013; X. Chen et al., 2013; Qiu et al., 2010). One study also found tangshenoside III (**12**) (Chen et al., 2010).



2	R_1=H	R_2=caffeoyl	R_3=H	R_4=H
3	R_1=H	R_2=H	R_3=H	R_4=caffeoyl
4	R_1=H	R_2=H	R_3=caffeoyl	R_4=H
5	R_1=caffeoyl	R_2=caffeoyl	R_3=H	R_4=H
6	R_1=caffeoyl	R_2=H	R_3=caffeoyl	R_4=H
7	R_1=caffeoyl	R_2=H	R_3=H	R_4=caffeoyl
8	R_1=H	R_2=H	R_3=caffeoyl	R_4=caffeoyl
9	R_1=caffeoyl	R_2=succinoyl	R_3=H	R_4=caffeoyl
10	R_1=caffeoyl	R_2=H	R_3=succinoyl	R_4=caffeoyl
11	R_1=caffeoyl	R_2=succinoyl	R_3=succinoyl	R_4=caffeoyl

Fig. 3. Structures of phenylpropanoids in *Saussurea involucrata.*

5.2 Flavonoids

Many studies have reported finding a variety of flavonoids in *Saussurea involucrata*. These different flavonoids can be categorized into four sub-groups, namely flavone aglycones, flavone glycosides, flavonol aglycones and flavonol glycosides.

The flavone aglycones include hispidulin (**13**), jaceosidin (**14**), luteolin (**15**), nepetin (**16**), apigenin (**17**) and 5,6-dihydroxy-7,8-dimethoxyflavone (**18**). The flavone glycosides include apigenin 7-*O*-glycoside (**19**), hispidulin 7-*O*-glucoside (**20**), luteolin7-*O*-glucoside (**21**), nepetin 7-*O*-glucoside (**22**), apeginin 7-*O*-glucuronide (**23**), chrysoeriol 7-*O*-glucuronide (**24**), chrysoeriol 7-*O*-glycoside (**25**) and chrysoeriol 7-*O*-rhamnoside (**26**) (Iwashina et al., 2010; Jing et al., 2013; Qiu et al., 2010).

Two flavonol aglycones have been found, quercetin (**27**) and kaempferol (**28**). The flavonol glycosides found include quercetin 3-*O*-rhamnoside (**29**), quercetin 3-*O*-rutinoside (rutin) (**30**), quercetin 3-*O*-glucoside (**31**), isorhamnetin 3-*O*-rutinoside (**32**) and kaempferol 7-*O*-glucopyranoside (**33**) (Iwashina et al., 2010).

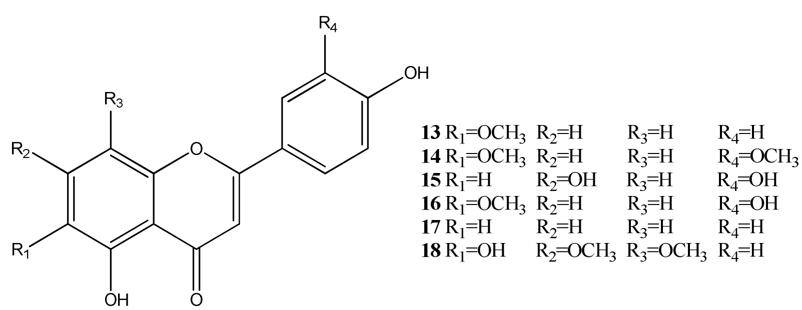


Fig. 4. Structures of flavone aglycones in *Saussurea involucrata.*

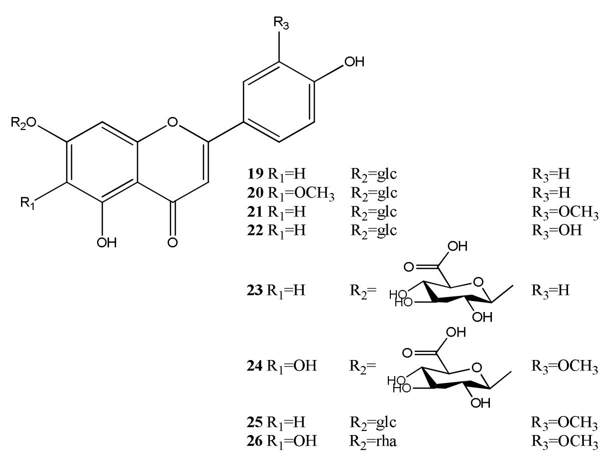
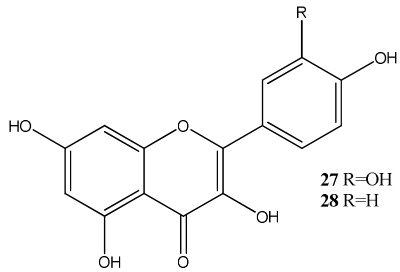
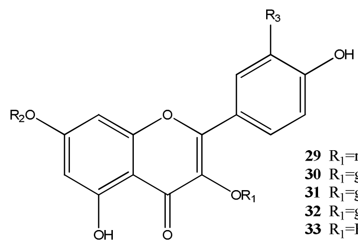


Fig. 5. Structures of flavone glycosides in *Saussurea involucrata.*



27 R=OH
28 R=H

Fig. 6. Structures of flavonol aglycones in *Saussurea involucrata.*

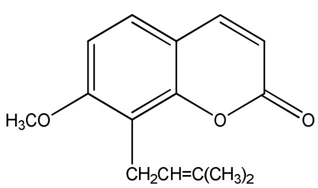


29	R_1=rha	R_2=H	R_3=OH
30	R_1=glc-glc	R_2=H	R_3=OH
31	R_1=glc	R_2=H	R_3=OH
32	R_1=glc-glc	R_2=H	R_3=OCH_3
33	R_1=H	R_2=glc	R_3=H

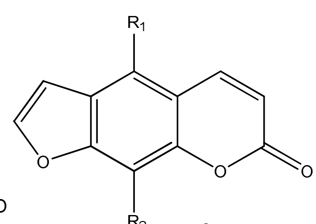
Fig. 7. Structures of flavonol glycosides in *Saussurea involucrata.*

5.3 Coumarins

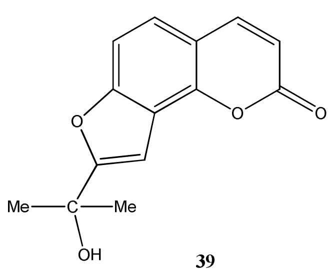
Eight coumarins have been isolated from *Saussurea involucrata*. These include one simple coumarin, osthol (**34**), and seven pyranocoumarins. The latter are isopimpinellin (**35**), bergapten (**36**), xanthotoxol (**37**), alloisoimperatorin (**38**), oroselol (**39**), edultin (**40**) and vaginidiol diacetate (**41**) (Yang et al., 2006).



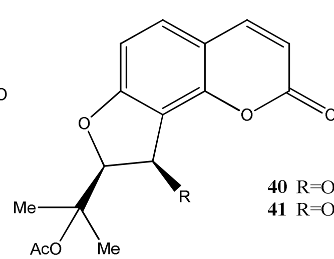
34



35	R_1=OCH_3	R_2=OCH_3
36	R_1=OCH_3	R_2=H
37	R_1=H	R_2=OH
38	R_1=OH	R_2=CH_2-CH=C(CH_3)$_2$



39



40 R=OCOC(CH_3)=CH(CH_3)
41 R=OAc

Figure 8 Structures of coumarins in *Saussurea involucrata.*

5.4 Lignans

Several lignans including arctigenin-4-*O*-(6''-*O*-acetyl-β-d-glucoside) (**42**), arctigenin-4-*O*-(2''-*O*-acetyl-β-d-glucoside) (**43**), arctigenin-4-*O*-(3''-*O*-acetyl-β-D-glucoside) (**44**), arctiin (**45**) and arctigenin (**46**) have been identified (Li et al., 2012; Liu and Aisa, 2010).

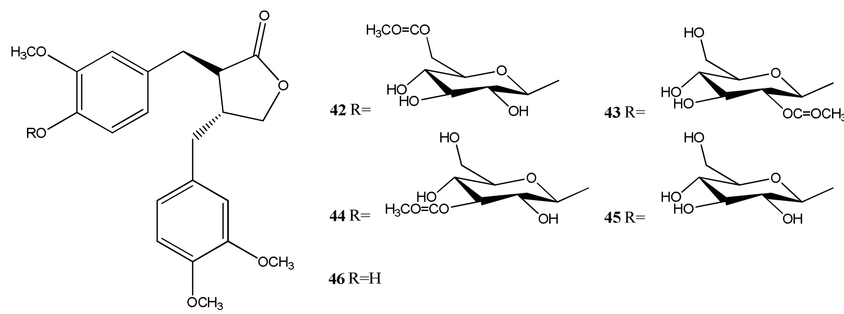


Fig. 9. Structures of lignans in *Saussurea involucrata.*

5.5 Sesquiterpenes

Sesquiterpenes are bitter substances. They often contain as a major structural feature an α,β-unsaturated-γ-lactone which, in recent studies, has been shown to be associated with anti-tumor, cytotoxic, anti-microbial and phytotoxic activities (Picman., 1983). Bioassay-directed separation of the ethyl acetate extract of the aerial parts of *Saussurea involucrata* has led to the isolation of sausinlactone A-(1S, 3S, 5S, 6S, 7S, 11S)-3-hydroxyl-11, 13-dihydrodehydrocostuslactone (**47**), sausinlactone B-(1S, 3S, 5S, 6S, 7S, 11R)-3-hydroxyl-11, 13-dihydrodehydrocostuslactone (**48**), sausinlactone C-(1S, 3S, 5S, 6S, 7S, 8S, 11S)-3-hydroxyl-11,13-dihydrodehydrocostuslactone (**49**), 3α, 8α-dihydroxyl-11βH-11, 13- dihydrodehydrocostuslactone (**50**) 8α-hydroxyl-

11βH-11,13-dihydrodehydrocostuslactone (**51**), 11β,13-dihydrodehydrocostuslactone-8-*O*-β-D-glucoside (**52**), 11β,13-dihydrodehydrocostuslactone-8-*O*-[6′-*O*-acetyl-β-D-glucoside] (**53**), 11α,13 -dihydroglucozaluzanin C (**54**) 3α-hydroxyl-11β,13-dihydrodehydrocostuslactone-8-*O*-β-D-glucoside (**55**) and japonicolactone (**56**) (Li et al., 2007; Wang et al., 2007). 10β,14-Dihydroxy-11βH-guai-4-(15)-ene-12,6α-olide 14-*O*-β-D-glucoside (**57**) and involucratin (**58**), and 11βH-2α-hydroxy-eudesman-4(15)-en-12,8β-olide, which is a eudesmanolide (**59**), were also isolated (Chen et al., 2010).

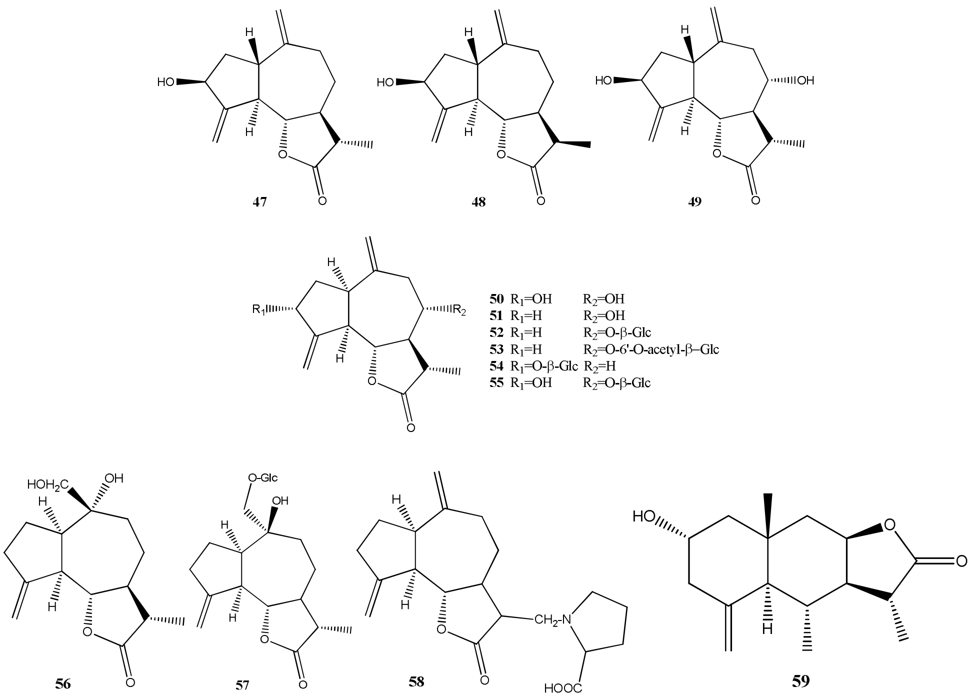


Fig. 10. Structures of sesquiterpenes in *Saussurea involucrata.*

5.6 Steroids

Seven steroids have been isolated from *Saussurea involucrata*. These are: bufotalin (**60**), telocinobufagin (**61**), gamabufotalin (**62**), daucosterol (**63**), β-sitosterol (**64**), 3-*O*-(6'-*O*-palmitoyl-β-D-glucosyl)-β-sitosterol (**65**) and 3-*O*-(6'-*O*-linoleoyl-β-D-glucosyl)- β-sitosterol (**66**) (Chen et al., 2010; Wu et al., 2009).

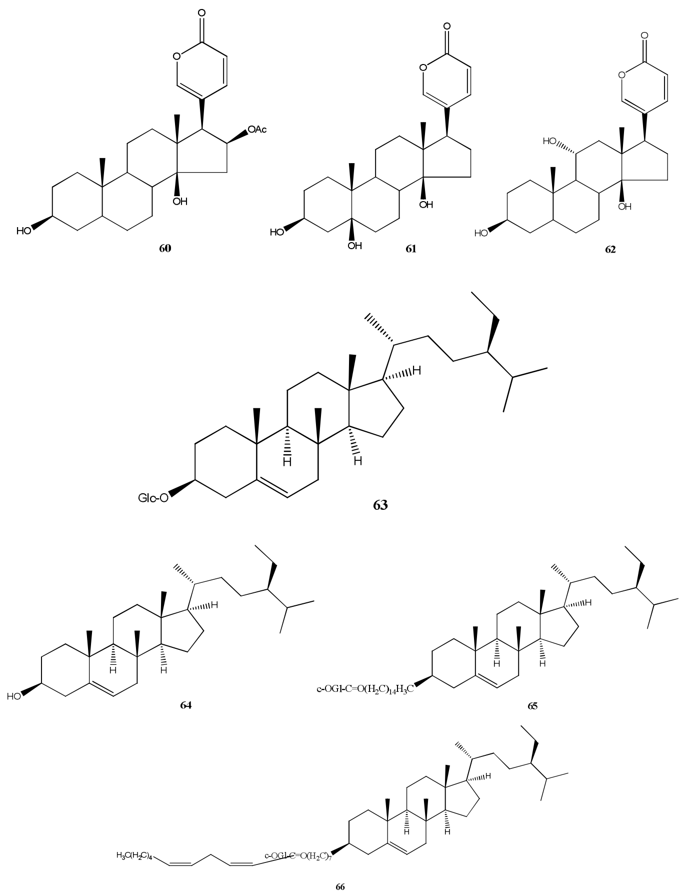


Fig. 11. Structures of steroids in *Saussurea involucrata*.

5.7 Ceramides

Rel-(3R,4S,5S)-3-[(2R)-2-hydroxynonadecanoyl~pentacosanoylamino]-4-hydroxy -5-[(4E)-heptadecane-4-ene]-2, 3, 4, 5-tetrahydrofuran (**67a-67g**) were the identified ceramides from *Saussurea involucrata* (Wu et al., 2009).

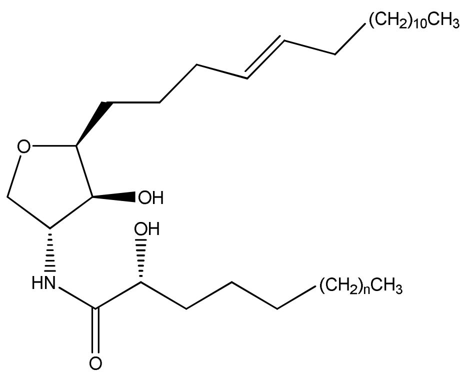


Fig. 12. Structures of ceramides in *Saussurea involucrata.*

5.8 Polysaccharides

Total polysaccharides with high purities have been separated from *Saussurea involucrata* through adsorption using macroporous resin and precipitation using ethanol (Wang et al., 2012). Polysaccharides from cultivated *Saussurea involucrata* (CSIP) were purified; two major fractions (CSIP1-2 and CSIP2-3) were investigated for their molecular weights, monosaccharide compositions and *in vitro* antioxidant activities. According to the results, the molecular weights of CSIP1-2 and CSIP2-3 were approximately 163.5 kDa and 88.6 kDa, respectively. CSIP1-2 was composed of glucose, galactose, xylose, rhamnose, arabinose and galacturonic acid with a molar ratio of 1.65: 0.39: 0.06: 8.33: 1.76: 40.43. CSIP2-3 was composed of glucose, galactose, xylose, rhamnose, arabinose and galacturonic acid with a molar ratio of 0.76: 0.66: 0.11: 5.59: 0.32: 44.66 (Yao et al., 2012).

6. Bioactivity

Saussurea involucrata is traditionally used to treat a wide spectrum of disorders; its efficacy is grounded by long history of usage and promising experimental results. Traditional clinical applications were considered guide for bioactivity evaluations, and many correlations were found between the pharmacological action in traditional records of the herb and bioactivities confirmed in modern studies (**Table 2**). Researches have revealed more bioactivities and its therapeutic spectrum of *Saussurea involucrata* is still broadening, with scientific evidence.

6.1 Anti-neoplastic

It has been an emerging clinical application for *Saussurea involucrata* to be used as an anti-neoplastic agent in the recent decade. *Saussurea involucrata* has gained widespread global praise owing to its effectiveness in anti-cancer therapy. Experiments have verified the anti-cancer bioactivity of *Saussurea involucrata* through different mechanism of action, including its interference on the cell growth process (Byambaragchaa et al., 2013), mainly by causing cell apoptosis (Chen et al., 2012; Way et al., 2010) and cytotoxicity (Wu et al., 2009; Xiao et al., 2011b; X. Zhang et al., 2011); affecting cell adhesion, migration and aggregation; while in gene level, it also has certain impact on transcription activity (Byambaragchaa et al., 2013). Human cancer cell lines were mainly targets in the anti-cancer bioactivities assessments and different assays were used to monitor the level of certain protein expression in the tumor cells (Byambaragchaa et al., 2013; Chen et al., 2012; Way et al., 2010; Wu et al., 2009; X. Zhang et al., 2011) while *in vivo* experiments involved the xenograft model (Chen et al., 2012; Way et al., 2010). The principal active compound has not yet been confirmed so far but experiments have suggested high possibility for it to be lying within the ethanol extract (75-95%) or the ethyl acetate fraction. The effective dose ranged from microgram level *in vitro* to milligram level *in vivo*; dose- and time-dependent manner has been demonstrated in some studies (Byambaragchaa et al., 2013; Chen et al., 2012; Way et al., 2010; Wu et al., 2009; X. Zhang et al., 2011).

6.2 Anti-arthritic

Rheumatoid arthritis (RA) is a chronic, inflammatory, systemic autoimmune disorder of withheld etiology for which there is no cure. (Doan and Massarotti, 2005) It is characterized by synovial inflammation and destruction of cartilage and bone (Klareskog et al., 2014). Currently, the aim of treatment is to mitigate the symptoms and prevent disability (Doan and Massarotti, 2005). *Saussurea involucrata*, which is used for the treatment of rheumatoid arthritis in folk medicines, has demonstrable anti-inflammatory and analgesic effects (Zhai et al., 2010).

The anti-inflammatory activity of *Saussurea involucrata* extract has been observed in mice and rats, with xylene or carrageenan-induced paw edema and ear edema (Jia et al., 2011; Wang et al., 2011; Yi et al., 2010; Zhai et al., 2010). Acetic acid-induced excessive abdominal capillary permeability (Jia et al., 2011; Zhai et al., 2010), primary and secondary adjuvant arthritis were the other models used in anti-inflammatory evaluation (Tao et al., 2007; Wang et al., 2011). RAW 264.7 macrophage were monitored for examination of the inhibitory effect of *Saussurea involucrata* in nitric oxide (NO) and prostaglandin E_2 (PGE_2) production in LPS-activated macrophage (Xiao et al., 2011a). NO is considered as a pro-inflammatory mediator that induces inflammation due to over production in abnormal situations (Sharma et al., 2007). PGE_2 is one of the most abundant PGs produced in the body. During inflammation, PGE_2 is of particular interest because it is involved in all processes leading to the classic signs of inflammation: redness, swelling and pain (Ricciotti and FitzGerald, 2011; Funk, 2001). Therefore, their relative abundance in the animal models can act as indicators for the degree of inflammation.

Investigations on the analgesic effects included acetic acid-induced writhing (Jia et al., 2011; Wang et al., 2011; Zhai et al., 2010), hot-plate test (Zhai et al., 2010) in mice. Central analgesic effect was also demonstrated by (Almeida and Navarro, 2001).

Ethanol extracts of *Saussurea involucrata* were given to mice and rats via intragastric route (*i.g.*) with dose ranging from 15-400 mg/kg and not more than 42 mg/kg in rats depending on the area of edema for no longer than 7 days (Jia et al., 2011; Wang et al., 2011; Yi et al., 2010); while a much lower dose of 0.36 mg/kg was given to mice by injection form (Tao et al., 2007). According to the report in the rat paw edema model (Yi et al., 2010), the peak inhibitory effects of *Saussurea involucrata* (42.2%) were recorded with a dose of 400 mg/kg at 3h post-carrageenan injection; while oral administration of *Saussurea involucrata* extract (400 mg/kg) resulted in a significant 33.3% inhibition of ear edema in mice. Flavonoids, found to be present in plasma after administration of the extracts, are believed to be the basis of the observed pharmacological effects (Yi et al., 2010).

In addition, the tissue culture of *Saussurea involucrata* was found to exhibit anti-inflammatory and analgesic activities, suggesting that cultured tissue of *Saussurea involucrata* could substitute for wild-grown plant material in the pharmaceutical industry (Jia et al., 2011).

However, the resemblance of animal models of arthritis to the patients developing symptoms and therefore the predictive power of these researches is in doubt. Animal models of arthritis are widely used to de-convolute disease pathways and to identify novel drug targets and therapeutic approaches. However, the high attrition rates of drugs in phase II/III rates means that a relatively small number of drugs reach the market, despite showing efficacy in pre-clinical models (McNamee et al., 2015). Other assays for better prediction of efficacy of herbs in treating arthritis are anticipated (McNamee et al., 2015).

6.3 Anti-oxidative and anti-aging

According to the free radical theory of aging, aging results from the accumulation of deleterious effects caused by free radicals, and the ability of an organism to cope with cellular damage induced by reactive oxygen species (ROS) plays an important role in determining organismal lifespan (Harman, 1956). Experimentally, increased ROS production is frequently detected in aged tissues (Maynard et al., 2009; Sawada et al., 1992; Sohal and Sohal, 1991) and many studies have found that increased oxidative damage in cells is associated with aging (Chakravarti and Chakravarti, 2007; Fraga et al., 1990; Hamilton et al., 2001; Oliver et al., 1987).

With reference to traditional records (Zhao, 1963), *Saussurea involucrata* has been used to postpone aging by invigorating *Yin* and *Yang* in body, which in some sense means promoting the activity of different organs in body. Multiple phytochemicals from *S. involcurata* were found to possess anti-oxidative pharmacological function. Polysaccharides of *Saussurea involucrata* have demonstrated anti-oxidative effect by scavenging superoxide anions in nitroblue tetraazolium colorimetric method, DPPH, hydroxyl and ABTS radical scavenging assay and by inhibiting the formation of thiobarbituric acid reactant in mouse liver homogenate (Yao et al., 2012; Zheng et al., 1993). Research has also revealed a positive correlation between antioxidant activity and the amount of phenolic and flavonoid compounds in *Saussurea involucrata* extracts, suggesting they could be the active constituents accounting for the antioxidant activity (Qiu et al., 2010; Wang et al., 2012). Further *in vitro* bioactive investigations have demonstrated that 3, 5-dicaffeoyl-1-*O*-(2-*O*-caffeoyl-4-maloyl)-quinic acid has significant anti-oxidative effect in DPPH and ABTS radical scavenging assays (Zou et al., 2014).

The oxidation-inhibitory effects were found to be in a concentration-dependent manner by comparison with the effects of the same doses of vitamin C that the scavenging effect of *Saussurea involucrata* on free radicals increased with quantity (Lee et al., 2011).

6.4 Anti-fatigue

Physiological fatigue, an incapacitating or disabling illness, means a reduction in the force output and energy generating capacity of a body after chronic exposure to work or usual activities at the same intensity (Shevchuk, 2007). Studies focused on the character of reactive oxygen species in confirmation of anti-fatigue bioactivity. Extensive evidence has accumulated demonstrating the beneficial effects of antioxidants in chronic fatigue. *Saussurea involucrata's* effectivenesss in coping with the ROS has been discussed in section 6.3. Besides, the ability of *Saussurea involucrata* to reduce fatigue has also been tested in experiments with mice. Decreased oxygen consumption and prolonged swimming time were observed after administration of polysaccharides of *Saussurea involucrata*. This provides clear evidence of its anti-fatigue effect and supports the traditional belief that the Snow Lotus herb can strengthen and invigorate the body (Zheng et al., 1993).

6.5 Neuro-protective and anti-hypoxic

Petrolleum ether extract, ethyl acetate extract and ethanol extract of *Saussurea involucrata* were found to possess neuro-protective effect and can reverse damage of brain cells under hypoxia state (Ma et al., 2014a, 2014b; Yang et al., 2011).

In hypoxic state, hypoxia-inducible factor-1 (HIF-1), which is a transcription factor and also a heterodimer composed of HIF-1α and HIF-1β protein subunits. It is essential for the activation of hypoxia-inducible genes like erythropoietin, some glucose transporters, the glycolytic enzymes, and vascular endothelial growth factor.

As HIF-1 activation may promote cell survival in hypoxic tissues, studies focused on the effect of hypoxic preconditioning on HIF-1 expression in hypoxia models of mice or rats (Bergeron et al., 2000; Ma et al., 2014b; Yang et al., 2011) by Western Blot assay (Ma et al., 2014b). Metabolism parameters, including ATP, ATPase, LAC, LDH, LD (Ma et al., 2011; Yang et al., 2011) and levels of oxidative stress indicators like SOD and MDA levels (Ma et al., 2014a; Yang et al., 2011) in pretreated hypoxic mice models were also determined in some studies. Besides, in other experiments, prolonged survival time of mice in conditions of acute anoxia and drop in mortality under acute decompression conditions were shown (Ma et al., 2011; Yang et al., 2011). Extracts had to be administered via intraperitoneal injection route (Ma et al., 2014a, 2014b) in order to be blood-brain-barrier crossing.

The neuroprotective activity of *Saussurea involucrata* has been demonstrated by the ability of its ethyl acetate fraction to inhibit MDA expression level, increase GPx activity, and decrease the expressions of COX-2, PARP and caspase-3, via downregulation of NF-kappaB. This biochemical cascade ultimately protects neural tissue (Y. L. Chen et al., 2013).

6.6 Immunomodulation

Both whole extracts (Jia and Wu, 2007) and specific components of *Saussurea involucrata* including flavones (Fan et al., 1996; Ma et al., 1998), XL-12 fraction of extraction (Wang et al., 2011) have been shown to have modulatory effects on immunity. Non-specific defense mechanisms and delayed hypersensitivity in mice has been inhibited by *Saussurea involucrata*, but humoral immunity activity has been stimulated upon administration of higher doses of *Saussurea involucrata* (Jia and Wu, 2007). Another experiment also reveals that *Saussurea involucrata* extract has an anti-allergic effect (Wang et al., 2011).

Enhanced activity of mononuclear phagocytic system was reflected by increased colloidal carbon clearance index rate demonstrated in an *in vivo* mice study (150 mg/kg; once a day; intragastrically adminstrated for a consectutive 7 days) (Fuller, 1992; Jia and Wu, 2007). *Saussurea involucrata* has also induced cytotoxicity of PBMC against K562 myelocytic leukemia cells and Raji Burkitt lymphoma cells and enhanced suboptimal concentration of rhIL-2-induced cytotoxicity but exhibited no effect on the large dose rhIL-2-induced cytotoxicity (Ma et al., 1998). Besides, significant enhancement of murine immunological function was observed, as reflected in increased percentage of lymphocyte transformation induced by PHA, the titer of serum hemagglutinin antibody against SRBC and hemolysin antibody (Fan et al., 1996). Apart from that, proliferation of T lymphocytes was promoted while B lymphocyte proliferation was hindered by *Saussurea involucrata* injection of the same concentration (Tao et al., 2007) and this serves as a ultimate proof of *Saussurea involucrata*'s peformance in two way adjustment function.

These research finding is consistent with Traditional Chinese Medicine usage as *Saussurea involucrata* has been described possessing invigorating power of both *Yin and Yang*, which include a meaning of maintaining balance in body.

6.7 Other pharmacological activities

It has been listed in Supplement to Compendium of Materia Medica (Zhao, 1963) since *Qing Dynasty*, which is a few hundred years ago, that *Saussurea involucrata* can be used to treat infertility due to aging (Zhao, 1963). Besides, *Saussurea involucrata* has been listed in modern Pharmacopoeia of China and local Pharmacopoeia regarding its menstrual cycle regulation effect (Chinese Pharmacopoeia Commission, 2010; Liu and Shawuti, 1985). Researches have consistently demonstrated its effect in prevention of premature ovarian failure of mice, which has correlations to estrogen regulation, caused by continuous light upon intra-gastric administration of 0.5 mL extract of *Saussurea involucrata* together with 0.9% of NaCl (Zhang et al., 2013).

Other bioactivities of *Saussurea involucrata* include anti-microbial and anti-fungal (Lv et al., 2010), anti-hypertensive (Yu and Chen, 2009), and anti-radiation (Gao et al., 2003; Jia et al., 2005). It appears that its water-soluble constituents, flavonoids and polysaccharides are responsible for these effects. (Chinese Pharmacopoeia Commission, 2010) Furthermore, *Saussurea involucrata* also has the ability to increase replication of bone mesenchymal stem cells (A. G. Zhang et al., 2011)

Table 2 Bioactivities of *Saussurea involucrata* as demonstrated in experiments.

Activity	Experimental protocol	Target animal(s)/ cell(s)	Result (+/-)	Dose	Chemical(s) /extract(s)	Reference(s)
Anti-neoplastic						
Cell growth	CCK-8 cell proliferation assay kit	SK-Hp1 human HCC cell line	+	200-400 μg/mL	95% ethanol extract	(Byambaragchaa et al., 2013)
> Cell cycle arrest	P21 and p27, cyclin D1, CDK4 biomarker	Human PC-3 cells	+	12.5-200 μg/mL	Ethyl acetate fraction of SI (incl. hispidulin, rutin)	(Chen et al., 2012; Way et al., 2010)
> Apoptosis	Bax expression, cytochrome C release, caspase-3 and caspase -9 activation, Bcl-2 expression	Human PC-3 cells	+	12.5-200 μg/mL	Ethyl acetate fraction of SI (incl. hispidulin, rutin)	(Chen et al., 2012; Way et al., 2010)
-	EGFR overexpression, Akt, STAT3	PC-3 xenograft model	+	dose-dependent; 10 and 30 mg/kg; i.g.; 3 times/week	Ethyl acetate fraction of SI (incl. hispidulin, rutin)	(Chen et al., 2012; Way et al., 2010)
> Cytotixicity	Cytotoxicity MTT assay	GepG2, MCF-7 cell lines	+	IC_{50} 0.05-0.5 μM	Bufotalin, telocinobufagin, gamabufotalin	(X. Zhang et al., 2011)
		HL-60, A375-S2, HeLa cell lines	+	-	Ceramides (75% ethanol extract)	(Wu et al., 2009)
		A549 cells	+	IC_{50} 0.01 μM (A) IC_{50} 2.89 μM (B)	Susquiterpene lactones sausinlactones A, B	(Xiao et al., 2011b)
Cell adhesion	Cell adhesion assay	SK-Hp1 human HCC cell line	+	200-400 μg/mL	95% Ethanol extract	(Byambaragchaa et al., 2013)
Cell migration	Wound closure assay	SK-Hp1 human HCC cell line	+	200 μg/mL	95% Ethanol extract	(Byambaragchaa et al., 2013)
Cell aggregation	Cell aggregation assay	SK-Hp1 human HCC cell line	+	400 μg/mL	95% Ethanol extract	(Byambaragchaa et al.,

						2013)
Gene transcriptional level and activity	mRNA analysis, reverse transciption-PCR, quantitative real time assays	SK-Hp1 human HCC cell line	+	dose-dependent	95% Ethanol extract	(Byambaragchaa et al., 2013)
Anti-arthritis related disorders						
Anti-inflammatory	Xylene induced-ear edema	Mice	+	-	30-50% Ethanol extract	(Zhai et al., 2010)
		Mice	+	15-60 mg/kg	XL-12	(Wang et al., 2011)
		Mice	+	-	95% Ethanol extract	(Jia et al., 2011)
	Xylene-induced paw edema	Mice	+	400 mg/kg; i.g.	50% Ethanol extract	(Yi et al., 2010)
	Carrageenan-induced paw edema	Rats	+	400 mg/kg; i.g.	50% Ethanol extract	(Yi et al., 2010)
	Carrageenan induced-hind paw edema	Rats	+	75-300 mg/kg; i.g.; 7 d	95% Ethanol extract	(Jia et al., 2011)
	Acetic acid-induced excessive abdominal capillary permeability	Mice	+	-	30-50% Ethanol extract	(Zhai et al., 2010)
		Mice	+	75-300 mg/kg; i.g.; 7 d	95% Ethanol extract	(Jia et al., 2011)
	Adjuvant arthritis induced by injection of Freud's complete adjuvant (primary)	Mice	+	0.36 mg/kg; inj.	-	(Tao et al., 2007)
		Rats	+	10.5-42 mg/kg	XL-12	(Wang et al., 2011)
	Adjuvant arthritis induced by injection of Freund's complete adjuvant (secondary)	Rats	+	10.5-42 mg/kg	XL-12	(Wang et al., 2011)
	RAW 264.7 macrophage proliferation, NO production	RAW 264.7 macrophage	+	25-200 μmol/L	95% Ethanol extract	(Xiao et al., 2011a)
Analgesic	Acetic acid induced-writhing test	Mice	+	-	30-50% Ethanol extract	(Zhai et al., 2010)
		Mice	+	75-300 mg/kg; i.g.;	95% Ethanol extract	(Jia et al., 2011)

				7 d		
		Mice	+	15-60 mg/kg	XL-12	(Wang et al., 2011)
	Hot plate test	Mice	+	-	30-50% ethanol extract	(Zhai et al., 2010)
	Central analgesic	Mice	+	20 mg/kg	NA	(Almeida and Navarro, 2001)
Immunomodulation [+ Stimulatory; - Inhibitory]						
	Carbon clearance rate(Non-specific immunity)	Mice	-	150 mg/kg; i.g.; 7 d	Ethanol extract	(Jia and Wu, 2007)
	Serum hemolysis formation	Mice	+	300 mg/kg; i.g.; 7 d	Ethanol extract	(Jia and Wu, 2007)
	Peripheral blood mononuclear cells (PBMC) specific cytotoxicity to Human K562, Raji cell lines	PBMC	+	0.6 μg/mL	Total flavone	(Ma et al., 1998)
	PHA induced-lymphocyte transformation percentage	Mice	+	40-80 μg/kg	Total flavone	(Fan et al., 1996)
	Serum hemagglutination antibody against SRBC	Mice	+	40-80 μg/kg	Total flavone	(Fan et al., 1996)
	Hemolytic antibody	Mice	+	40-80 μg/kg	Total flavone	(Fan et al., 1996)
	DNCB induced-delated hypersensitivity (non-specific immunity)	Mice	-	300 mg/kg; i.g.; 7d	Ethanol extract	(Jia and Wu, 2007)
	T cell and B lymphocyte proliferation	Rats	-	0.13-0.14 mg/mL	XL-12	(Wang et al., 2011)
Neuro-protective and anti-hypoxic						
	Metabolism parameters (ATP, ATPase, LAC, LDH)	Mice	+	51 mg/kg	Petroleum ether extract	(Ma et al., 2011)
	Metabolism parameters (LD, LDH, ATP, ATPase)	Normobaric, decompression, and chemistry poisoning	+	250-500 mg/kg	Ethanol extract	(Yang et al., 2011)

		hypoxia in mice				
		Brain tissues of hypoxia in rats	+	125-500 mg/kg; i.p.	Petroleum ether extract	(H. P. Ma et al., 2014a)
	Oxidative stress indicators (SOD and MDA levels)	Normobaric, decompression, and chemistry poisoning hypoxia in mice	+	250-500 mg/kg	Ethanol extract	(Yang et al., 2011)
		Brain tissues of hypoxia rats (hypoxia)	+	125-500 mg/kg; i.p.	Petroleum ether extract	(H. P. Ma et al., 2014a)
		D-galactose induced-brain injury in mice	+	30 mg/kg/d; 6 weeks	Ethyl acetate extract; rutin	(Y. L. Chen et al., 2013; Yang et al., 2012)
	Blood sugar, muscle glycogen and hepatic glycogen under chronic decompression conditions	Mice	+	52 mg/kg	Petroleum ether extract	(Ma et al., 2011)
	Survival time under hypoxia conditions	Normobaric, decompression, and chemistry poisoning hypoxia in mice	+	250-500 mg/kg	Ethanol extract	(Yang et al., 2011)
	Mortality under acute decompression conditions	Mice	+	50 mg/kg	Petrolleum ether extract	(Ma et al., 2011)
	Western Blot assay (HIF-1alpha gene expression)	Brain tissues of hypoxia in rats	+	125-500 mg/kg; i.p.	Petrolleum ether extract	(H. P. Ma et al., 2014b)
	Gene expression assay (EPO, HO-1 mRNA expression)	Brain tissues of hypoxia in rats	+	125-500 mg/kg; i.p.	Petrolleum ether extract	(H. P. Ma et al., 2014b)
Anti-oxidative and anti-aging						
	Superoxide radical assay	-	+	-	Polysaccharides	(Yao et al., 2012)
		-	+	-	Apigenin	(Fan and Yue, 2003; Li

						et al., 2006; Matkowski, 2008)
		-	+	10^{-6} M	Quercetin, rutin, morin	(Chen et al., 1990)
	DPPH radical scavenging assay	-	+	-	Polysaccharides	(Yao et al., 2012)
		-	+	16 mg/mL	Methanol extract	(Wang et al., 2012)
		-	+	0.74- 1.05 mmol/L	Phenolic acids	(Zou et al., 2014)
	Hydroxyl radical scavenging assay	-	+	-	Polysaccharides	(Yao et al., 2012)
	ABTS radical scavenging assay	-	+	0.94 mg/mL	Methanol extract	(Wang et al., 2012)
		-	+	54.75 μmol/g	PCA, CGA, rutin, cynarine, hispidulin	(Qiu et al., 2010); (Qiu et al., 2013)
		-	+	0.55 - 0.99 mmol/L	Phenolic acids	(Zou et al., 2014)
	Ferric reducing/antioxidant power (FRAP) assay	-	+	82.62 μmol/g	PCA, CGA, rutin, cynarine, hispidulin	(Qiu et al., 2010) (Qiu et al., 2013)
	Anti-aging Lipid peroxidation	D-galactose induced-brain injury in mice	+	30 mg/kg/d; 6 weeks	Ethyl acetate extract; rutin	(Yang et al., 2012)
Anti-fatigue						
	Reduction in serum urea nitrogen	Mice	+	1 g/kg; p.o.; 15 d	95% Ethanol extract	(Jia and Wu, 2008)
	Oxygen consumption and swimming time	Mice	+	-	Polysaccharides	(Zheng et al., 1993)
		Mice	+	1 g/kg; p.o.; 15 d	95% Ethanol extract	(Jia and Wu, 2008)

7. Toxicological studies

The use of *Saussurea involucrata* in pregnant women is prohibited in the Pharmacopoeia of China (Chinese Pharmacopoeia Commission, 2010). Similarly, it is proven to be abortion inducing in rats in early, mid-trimester and late pregnancy with different abortion rate. The highest rate has found to be as high as 100% in mid-trimester and late pregnancy for 0.3 mL administered intravenously once per day for consecutive two days. Early pregnancy is also sensitive to *Saussurea involucrata* injection. Apart from having toxicity to fetus, it is also accused for causing arrhythmia, hypotension, paralysis, nausea and other nervous and cardiovascular disorders as its adverse effects in overdose condition (Commission of Chinese Materia Medica, 1999).

However, contradictorily, *Saussurea involucrata* injection has been proven to be safe, with no evidence of any distinct toxicity or side effects in some researches. In one long-term toxicology test, rats were injected with *Saussurea involucrata* for 45 days; blood routine examination and pathologic histology examination showed no damaging effects (Sun et al., 2005). Another experimental study of the toxicity of long-term *Saussurea involucrata* injection was carried out. In this study, rats were given intraperitoneal injection for 90 days and were observed for 14 days subsequently. No abnormal changes of appearance or organ function, as indicated by blood and liver and kidney function evaluation, and pathological examination of 13 kinds of visceral tissue, were discovered. These studies, as well as the centuries of traditional use, confirm the safety of *Saussurea involucrata* in clinical use (Lian et al., 1996). Therefore, more qualified toxicological studies are expected for prediction of the therapeutic index so as to ensure safe use *Saussurea involucrata*.

8. Modern clinical application

8.1 Anti-arthritic

The uses of *Saussurea involucrata* are extensive (Xiao et al., 2011a); however, in modern actual practice, it is seldom used independently, but typically in association with other herbs nowadays.

Xuelian injection, which is the only pharmaceutical product that involved only extract of *Saussurea involucrata*, is officially used in hospitals in China for treatment of rheumatoid arthritis. Clinical trials were performed in order to evaluate its efficacy since 2006. One study involved 48 individuals who had developed rheumatoid arthritis with similar stage and 12 of them were randomly the positive control group which was injected with an efficacy-proven drug called 'Compound arthritis injection'. Xuelian injection of 4 mL was intramuscularly injected into the 36 individuals in experimental group once per day consecutively for 20 days. Total effective rate was found to be increased by 22.2% compared to the positive control group (Bao et al., 2006). Besides, another study has investigated the deviation in efficacy if the Xuelian injection was injected in more specific sites, i.e. acupuncture points according to the theory of Traditional Chinese Medicine. The rate of effectiveness was found to be increased to 100% in 60 tested individuals, with 4 mL of Xuelian injection dissolved in 2 to 3 mL of 0.9% NaCl injection once a day for 15 days. Reoccurrence was developed in only 25% of the individuals after one year of treatment (Gu et al., 2009).

8.2 Counteracting infertility

Recently, the efficacy of Xuelian injection has been tested on treating erectile dysfunction. Seventy individuals were involved and among them, thirty were randomly selected to be the positive control group with injection of Vitamin B with same dose as the experimental group while the remaining 40 individuals were injected, in specific acupuncture points, 0.5 mL of Xuelian injection. The result was promising, compared to the positive control group, an increase of 20.83% of effective rate, defined by successful vaginal penetration for not less than 1 min before ejaculation has been shown (Liu and Zhang, 2013).

8.3 Others application of *Saussurea involucrata*-containing compound prescription

As mentioned, compound prescription of herbs is one of the key characteristics of Traditional Chinese Medicine. The underlying principal is herbal interaction effects including synergistic and antagonistic effect. The chemical profile of an herb can be very sophisticated and interactions of herbs, to be more specific, of different chemical constituents in herbs, can further complicate the case. Hundreds of years of clinical practices in China have demonstrated that best efficacy is reached only when herbs are used together. Therefore, there are vast amount of pharmaceutical products containing, but not merely, *Saussurea involucrata* as the active components and they are used to treat a wide variety of disorders; these include neoplasm (Liang, 2013), cardiovascular diseases (Baima, 1997), allergies and asthma (Hu, 1996), dermatological disorders and injury (Xing et al., 2012).

This indicated the potential of *Saussurea involucrata* in treating a wide spectrum of diseases. Identification of principal active compound, however, would be of utmost importance for further studies on maximizing of efficacy while minimizing of toxicity.

9. Conclusion and prospects

Saussurea involucrata is a rare alpine herb that is frequently prescribed in various ethnomedical systems especially central Asia. This review summarized the existing botany, phytochemistry, pharmacological properties and application researches on *Saussurea involucrata*. The amount of modern experimental data manifested the multiple disease ameliorating properties of *Saussurea involucrata* and evidenced the traditional medical uses. Among them, the most widespread traditional use of this alpine herb has been for the treatment of inflammation. As inflammation has long been considered to be associated with the development of cancer, in recent decade, the emerging efficacy in treating cancer has further put *Saussurea involucrata* under the spotlight.

However, although increasing interest has prompted more studies on *Saussurea involucrata*, it is still noteworthy that several gaps in our understanding of its application exist. The first gap is that the intrinsically active compositions and the mechanism of action of *Saussurea involucrata* were ambiguous. For example, although *Saussurea involucrata* has gained widespread global praise owing to its effectiveness in anti-cancer therapy, the possible mechanism is still not conclusive and the responsible active components has not yet been confirmed only attributed to the ethanol extract or the ethyl acetate fraction. Therefore, further more studies should undoubtedly have the priority to identify the individual bioactive component and to more clearly dissect the molecular mechanism of the pharmacological effects of *Saussurea involucrata*.

Secondly, interestingly we have observed that the traditional use of *Saussurea involucrata* is seldom coupled with other herbs, while in modern clinical practice, *Saussurea involucrata* is usually used in associations with other herbs. Since compound prescription is one of the key characteristics of Traditional Chinese Medicine, therefore it is important to verify this contradictory phenomenon whether we should use this herb independently or in combination with other herbs by both traditional theory and modern pharmacological evidences.

Thirdly, the contents of chlorogenic acid and rutin, which have been set for the quality control of *Saussurea involucrata* in the Chinese Pharmacopoeia 2010 edition, varied from 1.19 to 14.43 mg/g and 0.14 to 16.02 mg/g respectively in the herbs obtained from different habitat and/or harvest's time. This may contribute to differences in the quality of various batches of *Saussurea involucrata*. Thus, how to exclusively and accurately monitor and evaluate quality of samples, to ensure and maintain their clinical and pharmaceutical stability, should be further studied. In addition, since it is proven to be abortion inducing in rats in early, mid-trimester and late pregnancy and also accused for causing arrhythmia, hypotension, paralysis, nausea and other nervous and cardiovascular disorders in overdose condition, further research into its toxic effects is necessary for prediction of the therapeutic index so as to ensure the safe use of *Saussurea involucrata*.

Lastly, *Saussurea involucrata*, under stress habitats in rocky and alpine environments, has limited distributions and grows very slowly. In recent years, the wild sources of *Saussurea involucrata* are dwindling dramatically due to the exhaustive exploitation. In order to cope with the problem of over-exploitation and its endangered species status, the use of *Saussurea involucrata* must be restricted to sustainable levels, and alternatives to the wild material must be developed. The use of the cultivated and/or tissue-cultured species as an alternative to the wild *Saussurea involucrata* should be encouraged, as studies have provided proof of their equivalent effectiveness. Another possibility is to explore other species of the genus *Saussurea* as potential substitutes for *Saussurea involucrata*. Other species are in fact currently being used, together with *Saussurea involucrata*, and recent research has demonstrated common biological activities and similarities in chemotaxonomy (Yi et al., 2009a; 2009b; 2010, 2012, 2014).

In a word, *Saussurea involucrata* is a valuable herb that is worth additional attention because of its wide uses, extensive biological activities, and reliable clinical efficacy. Deep phytochemical and pharmacological investigation of *Saussurea involucrata*, especially its mechanism of action, to illustrate its ethno-medicinal use will undoubtedly be the focus of future research.

Acknowledgments

This work was supported by the Faculty Research Grant of Hong Kong Baptist University (FRG2/14-15/061), the Natural Science Foundation of Guangdong Province (2014A030313766) and Science and Technology Fund of Sichuan Province for Outstanding Young Scholar (2012JQ0044).

Reference

Almeida, R.N., Navarro, D.S., Barbosa-Filho J.M., 2001. Plants with central analgesic activity. *Phytomedicine*. **8**, 310–322.

Baima, J.C., 1997. Tibetan preparatory medicine and preparation method thereof. Chinese Patent CN 1161849A.

Bao, L., Zhuo, Y., Chen, Z.C., Xu, X.Y., Li, X.M., 2006. Clinical trial of Xuelian injection in treatment of rheumatoid arthritis. *Chinese J. Inf. Tradit. Chinese Med*. **13**, 69–70.

Bergeron, M., Gidday, J.M., Yu, A.Y., Semenza, G.L., Ferriero, D.M., Sharp, F.R., 2000. Role of hypoxia-inducible factor-1 in hypoxia-induced ischemic tolerance in neonatal rat brain. *Ann. Neurol.* **48**, 285–296.

Byambaragchaa, M., de la Cruz, J., Yang, S.H., Hwang SG., 2013. Anti-metastatic potential of ethanol extract of *Saussurea involucrata* against hepatic cancer in vitro. *Asian Pac. J. Cancer Prev*. **14**, 5397–5402.

Chakravarti, B., Chakravarti, D.N., 2007. Oxidative modification of proteins: age-related changes. *Gerontology*. **53**, 128–139.

Chen, F.J., Yang, Y.G., Zhao, D.X., Gui, Y.L., Guo, Z.C., 1999. Advances in studies of species, habitats distribution and chemical composition of Snow Lotuses (*Saussurea*) in China. *Chinese Bull. Bot.* **16**, 561–566.

Chen, Q.L., Yi, T., Tang, Y.N., Wong, L.L., Huang, X.X., Zhao, Z.Z., Chen, H.B., 2014a. Comparative authentication of three “Snow Lotus” herbs by macroscopic and microscopic features. *Microsc. Res. Tech.* **77**, 631–641.

Chen, R.D., Liu, X., Zou, J.H., Yang, L., Dai, J.G., 2014b. Regulation of syringin, chlorogenic acid and 1, 5-dicaffeoylquinic acid biosynthesis in cell suspension cultures of *Saussurea involucrata*. *China J. Chinese Mater. Medica*. **39**, 2275–2280.

Chen, R.D., Zou, J.H., Jia, J.M., Dai, J.G., 2010. Chemical constituents from the cell cultures of *Saussurea involucrata*. *J. Asian Nat. Prod. Res*. **12**, 119–123.

Chen, R., Liu, X., Zou, J., Yang, L., Dai, J., 2013. Qualitative and quantitative analysis of phenylpropanoids in cell culture, regenerated plantlets and herbs of *Saussurea involucrata*. *J. Pharm. Biomed. Anal*. **74**, 39–46.

Chen, X.J., Liu, Q.H., Yu, P., Zhang, J., 2013. HPLC determination of chlorogenic acid and rutin in *Saussurea involucrata* (Kar. et Kir.) seed. *Xinjiang Med. J*. (9), 139–142.

Chen, Y., Hsiao, P., Lin, F., Wu, G., Tsai, C., 2012. Evidence-based anticancer complementary and alternative medicine, in Materia Medica for Various Cancers, Cho, William C.S. (Ed.). Springer Netherlands, Dordrecht, pp. 110–111.

Chen, Y.L., Tsay, H.S., Way, T. D., Lee, P.L., Yu, C.Y., Chen, C.H., 2013. *Saussurea involucrata* extract, pharmaceutical composition and use for anti-fatigue and anti-aging. United States Patent US 20130177661 A1.

Chen, Y.R., 1963. Medical dictionary of China. Jianwen Publisher, Hong Kong, pp. 1263.

Chen, Y.R., Wang, X.J., Aidarhan, N., Zhao, B., Yuan, X.F., Li, Y., 2013. Effects of mowing strength on the chemical composition content in *Saussurea involucrata* Kar. Et Kir. *Adv. Mater. Res*. **610-613**, 3432–3436.

Chen, Y.T., Zheng, R.L., Jia, Z.J., Ju, Y., 1990. Flavonoids as superoxide scavengers and antioxidants. *Free Radic. Biol. Med*. **9**, 19–21.

Chinese Medicinal Materials Corporation, 1994. Zhongguo Minjian Dan Yanfang. China Social Sciences Publishing House, Beijing, pp. 804.

Chinese Pharmacopoeia Commission, 2010. Pharmacopeia of the People’s Republic of China, volume **1**. Chemical Industry Press, Beijing, pp 50–51.

Commission of Chinese Materia Medica, 1999. Materia Medica of China, volume **7**. Shanghai Scientific and Technical Publishers, Shanghai, pp 927–929.

Commission of Chinese Materia Medica, 2005. Materia Medica of China (Uyghur medicine), volume **33**. Shanghai Scientific and Technical Publishers, Shanghai, pp. 338–339.

Committee for National Revolution of Xinjiang Uyghur Autonomous Region Health Bureau, 1976. Xinjiang Zhongcaoyao. Xinjiang People's Publishing House, Xinjiang, pp. 342–343.

Doan, T., Massarotti, E., 2005. Rheumatoid arthritis: an overview of new and emerging therapies. *J. Clin. Pharmacol*. **45**, 751–762.

Fan, C.Q, Yue, J.M, 2003. Biologically active phenols from *Saussurea medusa*. *Bioorg. Med. Chem*. **11**, 703–708.

Fan, C.Q., Zhu, X.Z., Zhan, Z.J., Ji, X.Q., Li, H., Yue, J.M., 2006. Lignans from *Saussurea conica* and their NO production suppressing activity. *Planta Med*. **72**, 590–595.

Fan, G.X., Ren, H.X., Yuan, Y.K., Liu, H.F., 1996. Effect of *Saussurea involucrata* Kar Et Kir Ex maxim flavone on murine immunology function. *J. Xi'An Med. Univ*. **17**, 452–454.

Flora of China Editorial Committee, 1999. Flora of China, volume **78**. Science Press, Beijing, China. pp 35–37.

Fraga, C.G., Shigenaga, M.K., Park, J.W., Degan, P., Ames, B.N., 1990. Oxidative damage to DNA during aging: 8-hydroxy-2'-deoxyguanosine in rat organ DNA and urine. *Proc. Natl. Acad. Sci. U. S. A*. **87**, 4533–4537.

Fu, C.X., Xu, Y.J., Zhao, D.X., Ma, F.S., 2006. A comparison between hairy root cultures and wild plants of *Saussurea involucrata* in phenylpropanoids production. *Plant Cell Rep*. **24**, 750–754.

Fu, L.G., Jin, J.M., 1991. China plant red data book: Rare and endangered plants. volume **1**. Science Press, Shanghai, China, pp. 234–235.

Fuller, R., 1992. Probiotics: The scientific basis. Springer Netherlands, Dordrecht.

Funk, C.D., 2001. Prostaglandins and leukotrienes : advances in eicosanoid biology. *Science*. **294**, 1871–1875.

Gao, B., Liang, Z.Q., Gu, Z.L., 2003. Protective effect of extract of *Saussurea involucrata* on radiation injured mice. *Chinese Tradit. Herb. Drugs*. **34**, 443–445.

Gu, Y., Xie, J.P., Yang, Y., 2009. Xuelian injection in acupuncture points in treating rheumatoid arthritis: 60 cases. *Shanxi J. Tradit. Chinese Med.* **25**, 30.

Guo, B., Abbasi, B.H., Wei, Y.H., 2011. Effects of hypobaric growth conditions on morphogenic potential and antioxidative enzyme activities in *Saussurea involucrata*. *Biol. Plant.* **55**, 783–787.

Guo, B., Gao, M., Liu, C.Z., 2007. *In vitro* propagation of an endangered medicinal plant *Saussurea involucrata* Kar. et Kir. *Plant Cell Rep.* **26**, 261–265.

Hamilton, M.L., Van Remmen, H., Drake, J.A., Yang, H., Guo, Z.M., Kewitt, K., Walter, C.A., Richardson, A., 2001. Does oxidative damage to DNA increase with age? *Proc. Natl. Acad. Sci. U. S. A.* **98**, 10469–10474.

Harman, D., 1956. Aging: a theory based on free radical and radiation chemistry. *J. Gerontol.* **11**, 298–300.

Hu, C.G., 1996. Chinese medicinal composition for treatment of trachitis and gastritis. Chinese Patent CN 1129134A.

Iwashina, T., Smirnov, S. V, Damdinsuren, O., Kondo, K., 2010. *Saussurea* species from the Altai Mountains and adjacent area, and their flavonoid diversity. *Bull. Natl. Mus. Nat. Sci., Ser. B,* **36**, 141–154.

Jia, J.M., Wu, C.F., 2007. Effect of *Saussurea involucrata* culture on mouse immune function. *China J. Tradit. Chinese Med. Pharm.* (4), 238–240.

Jia, J.M., Wu, C.F., 2008. Antifatigue activity of tissue culture extracts of *Saussurea involucrata*. *Pharm. Biol.* **46**, 433–436.

Jia, J.M., Wu, C.F., Liu, W., Yu, H., Hao, Y., Zheng, J.H., Ji, Y.R., 2011. Antiinflammatory and analgesic activities of the tissue culture of *Saussurea involucrata*. *Biol. Pharm. Bull.* **28**, 1612–1614.

Jia, J.M., Wu, C.F., Yu, H., Hu, G.S., 2005. Anti-radiation activity of the tissue culture of *Saussurea involucrata* Kar. et Kir. *J. Shenyang Pharm. Univ.* (6), 444–448, 462.

Jia, M.R., Li, X.W., Zhang H., Yan, Z.Y., Liao, X.R., 2005. Ethnomedicines in China (Zhongguo Minzuyao Zhiyao). China Medical Science Press, Beijing, pp. 544–545.

Jing, L.L., Fan, X.F., Fan, P.C., He, L., Jia, Z.P., 2013. 5,6-Dihy-droxy-7,8-di-meth-oxy-flavone. *Acta Crystallogr. Sect. E. Struct. Rep. Online*. **69**, o1096.

Kang, X.L., Wang, X.J., Li, T.N., Yuan, Y.X., 2010. Study on seed germination characteristics of the endangered & medicinal plant *Saussurea involucrata* Kar.et Kir. *Seed*. (5), 81–83.

Klareskog, L., Amara, K., Malmström, V., 2014. Adaptive immunity in rheumatoid arthritis: anticitrulline and other antibodies in the pathogenesis of rheumatoid arthritis. *Curr. Opin. Rheumatol*. **26**, 72–79.

Lee, J.C., Kao, J.Y., Kuo, D.H., Liao, C.F., Huang, C.H., Fan, L.L., Way, T.D., 2011. Antifatigue and antioxidant activity of alcoholic extract from *Saussurea involucrata*. *J. Tradit. Complement. Med*. **1**, 64–68.

Li, F.X., Jin, Z.P., Zhao, D.X., Cheng, L.Q., Fu, C.X., Ma, F.S., 2006. Overexpression of the *Saussurea medusa* chalcone isomerase gene in *S. involucrata* hairy root cultures enhances their biosynthesis of apigenin. *Phytochemistry*. **67**, 553–560.

Li, J.R., Liu, L., Liu, Y.Q., Sa, A.J., 2012. Determination of arctiin in seeds of *Saussurea involucrata* by HPLC. *Lishizheng Med. Mater. Medica Res*. **23**, 2119–2120.

Li, Y., Jia, Z.J., 1989. Guaianolides from *Saussurea involucrata*. *Phytochemistry*. **28**, 3395–3397.

Li, Y., Wang, C.L., Guo, S.X., Yang, J.S., Xiao, P.G., 2007. Three guaianolides from *Saussurea involucrata* and their contents determination by HPLC. *J. Pharm. Biomed. Anal*. **44**, 288–292.

Li, Y.M., Zhong, H., 2013. Simultaneous determination of five flavonoids in *Saussurea involucrata* by capillary electrophoresis. *J. Chem. Soc. Pakistan*. **35**, 1288–1292.

Lian, J., Xiong, Y.J., Zhu, H.H., Lu, X.M., 1996. Experimental study on long-term toxicity of Xuelian injection. *Mod. J. Integr. Tradit. Chinese West. Med*. **16**, 458–460.

Liang, S.J., 2013. Traditional Chinese medicine powder prepared from three flowers, five seeds and six whites for treating lung cancer. Chinese Patent CN 103432401A.

Liu, L.J., Zhang, J.J., 2013. Xuelian injection in treating erectile dysfunction. *Tradit. Chinese Med. Res*. **26**, 51–52.

Liu, Y.D., Aisa, H.A., 2010. Three new lignans from the seeds of *Saussurea involucrata*. *J. Asian Nat. Prod. Res*. **12**, 828–833.

Liu, Y.M., Shawuti, Y., 1985. Pharmacography of Uighur. Part 1. Xinjiang People's Publishing House, Urumqi, China, pp. 385–389.

Lv, Y.L., Zhang, F.S., Chen, J., Cui, J.L., Xing, Y.M., Li, X.D., Guo, S.X., 2010. Diversity and antimicrobial activity of endophytic fungi associated with the alpine plant *Saussurea involucrata*. *Biol. Pharm. Bull*. **33**, 1300–1306.

Madhavi, M., Mallika, G., Lokanath, N., Vishnu, M.N., Madhusudhana Chetty, C., Mohamed Saleem, T.S., 2012. A review on phytochemical and pharmacological aspects of *Saussurea lappa*. *Int. J. Life Sci. Med. Res*. **2**, 24–31.

Ma, H.J., Fan, G.X., Ren, H.X., Yuan, Y.K., 1998. Effect of *Saussurea involucrata* Kar Et Kir Ex Maxim flavone on human peripheral blood mononuclear cells. *J. Xi'An Med. Univ*. **19**, 168–169, 200.

Ma, H.P., Fan, P.C., Jing, L.L., Yao, J., He, X.R., Yang, Y., Chen, K.M., Jia, Z.P., 2011. Anti-hypoxic activity at simulated high altitude was isolated in petroleum ether extract of *Saussurea involucrata*. *J. Ethnopharmacol*. **137**, 1510–1515.

Ma, H.P., Jiao, Y., Gao, R.M., Yang, Y., Fan, P.C., Jing, L.L., Jia, Z.P., 2014a. Effect of ethanol extract from *Saussurea involucrata* on biochemical indicators of stimulated high-altitude hypoxia induced mice. *J. Chinese Med. Mater*. **37**, 99–103.

Ma, H.P., Yao, J., Wu, J.H., Gao, R.M., Fan, P.C., Jing, L.L., 2014b. Protective effect and action mechanism of petroleum ether extracts from *Saussurea involucrata* on brain tissues of hypoxia rats. *China J. Chinese Mater. Medica.* **39**, 2710–2715.

Ma, Q.D., Wang, X.Y., Bolati, M., 2012. Method for separation and enrichment of total flavones and total polysaccharides in *Saussurea involucrata* extract. Chinese Patent CN 201210247167.9.

Matkowski, A., 2008. Plant *in vitro* culture for the production of antioxidants--a review. *Biotechnol. Adv*. **26**, 548–560.

Maynard, S., Schurman, S.H., Harboe, C., de Souza-Pinto, N.C., Bohr, V.A., 2009. Base excision repair of oxidative DNA damage and association with cancer and aging. *Carcinogenesis.* **30**, 2–10.

McNamee, K., Williams, R., Seed, M., 2015. Animal models of rheumatoid arthritis: How informative are they? *Eur. J. Pharmacol*. **759**, 278–286.

Mpala, L., Cock, I.E., Chikowe, G., 2010. No evidence of antiseptic properties and low toxicity of selected Aloe species. *J. Pharm. Negat. Results*. **1**, 10–16.

National Institutes for Food and Drug Control, 1984. Zhongguo Minzu Yaozhi. People's Medical Publishing House, Beijing, pp. 448–449.

Oliver, C.N., Ahn, B.W., Moerman, E.J., Goldstein, S., Stadtman, E.R., 1987. Age-related changes in oxidized proteins. *J. Biol. Chem*. **262**, 5488–5491.

Picman, A.K., Towers, G.H.N. 1983. Antibacterial activity of sesquiterpene lactones. *Biochem. Syst. Ecol.*, **11**, 321-327.

Qiu, J., Gao, F., Shen, G., Li, C., Han, X., Zhao, Q., Zhao, D., Hua, X., Pang, Y., 2013. Metabolic engineering of the phenylpropanoid pathway enhances the antioxidant capacity of *Saussurea involucrata*. *PLoS One*. **8**, e70665.

Qiu, J., Xue, X., Chen, F., Li, C., Bolat, N., Wang, X., Baima, Y., Zhao, Q., Zhao, D., Ma, F., 2010. Quality evaluation of snow lotus (*Saussurea*): quantitative chemical analysis and antioxidant activity assessment. *Plant Cell Rep*. **29**, 1325–1337.

Ricciotti, E., FitzGerald, G.A., 2011. Prostaglandins and inflammation. *Arterioscler. Thromb. Vasc. Biol*. **31**, 986–1000.

Rivera, D., Allkin, R., Obón, C., Alcaraz, F., Verpoorte, R., Heinrich, M., 2014. What is in a name? The need for accurate scientific nomenclature for plants. *J. Ethnopharmacol*. **152**, 393-402.

Sawada, M., Sester, U., Carlson, J.C., 1992. Superoxide radical formation and associated biochemical alterations in the plasma membrane of brain, heart, and liver during the lifetime of the rat. *J. Cell Biochem*. **48**, 296–304.

Sharma, J.N., Al-Omran, A., Parvathy, S.S., 2007. Role of nitric oxide in inflammatory diseases. *Inflammopharmacology*. **15**, 252–259.

Shevchuk, N.A., 2007. Possible use of repeated cold stress for reducing fatigue in chronic fatigue syndrome: a hypothesis. *Behav. Brain Funct*. **3**, 55.

Sohal, R.S., Sohal, B.H., 1991. Hydrogen peroxide release by mitochondria increases during aging. *Mech. Ageing Dev*. **57**, 187–202.

Sun, Y.H., Hamrati, Zhang, Y.C., Yang, L.L., Li, T., Liu, F., 2005. Long-term toxicology study on *Saussurea involucrata* injection. *Lishizhen Med. Mater. Medica Res*. (2), 70–71.

Tao, H.Y., Huang, H., Hou, G.P., Liu, F., 2007. Effect of *Saussurea involucrata* injection on counteracting adjuvant-induced arthritis and its immonuregulation function. *Tradit. Chinese Drug Res. Clin. Pharmacol*. **18**, 269–273.

The Plant List, Version 1.1, 2013. URL http://www.theplantlist.org.

Wang, L.L., Wang, X., Liu, Y., Hua, Y., Yang, Q.L., Xu, F., Huang, H., 2011. Main pharmacodynamics of *Saussurea involucrata* extract XL-12. *Chinese Tradit. Pat. Med*. **33**, 1868–1874.

Wang, Q., Liu, L., Aisa, H.A., 2012. Chemical composition and antioxidant activity of *Saussurea involucrata* seed. *Chem. Nat. Compd*. **48**, 663–665.

Wang, X.L., Gesang, S.L., Jiao, W., Liao, X., Ding, L.S., 2007. Two new sesquiterpenoid glucosides from the aerial parts of *Saussurea involucrata*. *J. Integr. Plant Biol*. **49**, 609–614.

Wang Y.F., Ni, Z.Y., Dong, M., Cong, B., Shi, Q.W., Gu, Y.C., Kiyota, H., 2010. Secondary metabolites of plants from the genus *Saussurea*: chemistry and biological activity. *Chem. Biodivers*. **7**, 2623–2659.

Wang, Y.F., Wang, X.C., Xue, G.P., Xing, J.G., Ma, J.H., Ma, X.L., 2012. Preparation method of transdermal composition containing rutin and chlorogenic acid extracted from Chinese medicine *Saussurea involucrata*. Chinese Patent CN 102349938A.

Way, T.D., Lee, J.C., Kuo, D.H., Fan, L.L., Huang, C.H., Lin, H.Y., Shieh, P.C., Kuo, P.T., Liao, C.F., Liu, H.S., Kao, J.Y., 2010. Inhibition of epidermal growth factor receptor signaling by *Saussurea involucrata*, a rare traditional Chinese medicinal herb, in human hormone-resistant prostate cancer PC-3 cells. *J. Agric. Food Chem*. **58**, 3356–3365.

Wu, L., Guo, S., 2008. Interaction between an isolate of dark-septate fungi and its host plant *Saussurea involucrata*. *Mycorrhiza*. **18**, 79–85.

Wu, L.Q., Lv, Y.L., Meng, Z.X., Chen, J., Guo, S.X., 2010. The promoting role of an isolate of dark-septate fungus on its host plant *Saussurea involucrata* Kar. et Kir. *Mycorrhiza*. **20**, 127–135.

Wu, W., Qu, Y., Gao, H.Y., Yang, J.Y., Xu, J.G., Wu, L.J., 2009. Novel ceramides from aerial parts of *Saussurea involucrata* Kar. et. Kir. *Arch. Pharm. Res*. **32**, 1221–1225.

Xiao, W., Li, N., Bolati, M., Wang, X.J., Jia, X.G., Zhao, Y.Q., 2011a. Advances in studies on chemical components and pharmacological activities of Saussurea Involucratae Herba. *Drugs Clin*. **26**, 344–348.

Xiao, W., Li, X., Li, N., Bolati, M., Wang, X.J., Jia, X.G., Zhao, Y.Q., 2011b. Sesquiterpene lactones from *Saussurea involucrata*. *Fitoterapia.* **82**, 983–987.

Xie, J.S., 1968. Xibei Yuji. Yee Wen Publishing Company, Taibei.

Xie, Z.W., 1975. National Herbal Compendium. People’s Medical Publishing House, Beijing, pp. 863–864.

Xu, Y.J., Zhao, D.X., Fu, C.X., Cheng, L.Q., Wang, N.F., Han, L.J., Ma, F.S., 2009. Determination of flavonoid compounds from *Saussurea involucrata* by liquid chromatography electrospray ionization mass spectrometry. *Nat. Prod. Res*. **23**, 1689–1698.

Yang, J.S., Xie, F.Z., Liu, Q.H., Wu, X., 2006. Studies on coumarins of *Saussurea involucrata* Kar. et Kir. *Chinese Pharm. J*. **41**, 1774–1776.

Yang, Y., Ma, H.P., Chen, Y., Zhang, R.X., He, X.R., Yao, J., Fan, P.C., Jia, Z.P., 2011. Anti-hypoxia effects of ethanol extract from *Saussurea involucrata*. *Chinese J. Exp. Tradit. Med. Formulae.* **17**, 145–148.

Yang, Y.C., Lin, H.Y., Su, K.Y., Chen, C.H., Yu, Y.L., Lin, C.C., Yu, S.L., Yan, H.Y., Su, K.J., Chen, Y.L.S., 2012. Rutin, a flavonoid that is a main component of *Saussurea involucrata*, attenuates the senescence effect in D-galactose aging mouse model. *Evid. Based. Complement. Alternat. Med*. **2012**, 980276.

Yao, L., Zhao, Q., Xiao, J., Sun, J., Yuan, X., Zhao, B., Su, H., Niu, S., 2012. Composition and antioxidant activity of the polysaccharides from cultivated *Saussurea involucrata*. *Int. J. Biol. Macromol*. **50**, 849–853.

Yi, T., Chen, H.B., Zhao, Z.Z., Jiang, Z.H., Cai, S.Q., Wang, T.M., 2009a. Comparative analysis of the major constituents in the traditional Tibetan medicinal plants *Saussurea laniceps* and *S. medusa* by LC–DAD–MS. *Chromatographia.* **70**, 957–962.

Yi, T., Chen, H.B., Zhao, Z.Z., Jiang, Z.H., Cai, S.Q., Wang, T.M., 2009b. Identification and determination of the major constituents in the traditional Uighur medicinal plant *Saussurea involucrata* by LC-DAD-MS. *Chromatographia*. **69**, 537–542.

Yi, T., Lo, H.W., Zhao, Z.Z., Yu, Z.L., Yang, Z.J., Chen, H.B., 2012. Comparison of the chemical composition and pharmacological effects of the aqueous and ethanolic extracts from a Tibetan "Snow Lotus" (*Saussurea laniceps*) herb. *Molecules.* **17**, 7183–7194.

Yi, T., Zhao, Z.Z., Yu, Z.L., Chen, H.B., 2010. Comparison of the anti-inflammatory and anti-nociceptive effects of three medicinal plants known as "Snow Lotus" herb in traditional Uighur and Tibetan medicines. *J. Ethnopharmacol*. **128**, 405–411.

Yi, T., Zhu, L., Tang, Y.N., Zhang, J.Y., Liang, Z.T., Xu, J., Zhao, Z.Z., Yu, Z.L., Bian, Z.X., Yang, Z.J., Chen, H.B., 2014. An integrated strategy based on UPLC-DAD-QTOF-MS for metabolism and pharmacokinetic studies of herbal medicines: Tibetan "Snow Lotus" herb (*Saussurea laniceps*), a case study. *J Ethnopharmacol.* **153**, 701–713.

Yu, X.D., Chen, H., 2009. Effect of four species of characteristic vegetable drugs in Xinjiang on cardiovascular system. *LiShiZhen Med. Mater. Medica Res.* **20**, 1357–1358.

Zhai, K.F., Duan, H., Xing, J.G., Huang, H., 2010. Study on the anti-inflammatory and analgesic effects of various purified parts from *Saussurea involucrata*. *Chinese J. Hosp. Pharm.* (5), 374–377.

Zhang, A.G., Cai, J.P., Xu, B.M., Tan, X.L., 2011. Effect of four traditional Chinese medicine decoctions and ethanolic extract from *Saussurea involucrata* on bone marrow mesenchymal stem cell proliferation activity *in vitro*. *J. Clin. Rehabil. Tissue Eng. Res.* **15**, 4287–4290.

Zhang, J., Zhang, M.M., Wang, J., Zhang, B.B., Yang, H., Zhuang, S.Z., Zhang, X.H., 2013. Preliminary study on the *Saussurea involucrata* extraction in preventing premature ovariall failure of mice. *J. Anhui Agric. Sci.* **41**, 10622–10623, 10656.

Zhang, X., Ye, M., Dong, Y.H., Hu, H.B., Tao, S.J., Yin, J., Guo, D.A., 2011. Biotransformation of bufadienolides by cell suspension cultures of *Saussurea involucrata*. *Phytochemistry*. **72**, 1779–1785.

Zhao, X.M., 1963. Supplement to Compendium of Materia Medica. People’s Medical Publishing House, Beijing, pp. 249–251.

Zheng, R.L., Liu, G.S., Xing, G.X., Jia, Z.J., Du, M., Tan, L.Q., 1993. Free radical scavenging and anti-fatigue activities of *Saussurea involucrata* polysaccharides. *Acta Pharmacol. Sin.* **14**, S47–49.

Zou, X.W., Liu, D., Liu, Y.P., Fu, Y.H., Zhang, X.Z., Xiu, Z.L., Xiao, H.B., 2014. Isolation and characterization of two new phenolic acids from cultured cells of *Saussurea involucrata*. *Phytochem. Lett.* **7**, 133–136.