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Original Research

Effect of *Carthamus tinctorius* L. (Safflower) on National Institute of Health Stroke Scale Scores of Ischemic Stroke Patients: A Pilot Clinical Trial

Mahdi Yousefi[1], Payam Sasannezhad[2], Hassan Rakhshande[3], Hassan Doosti[4], Azade Saki[5], Zahra Baghestani Kouzegar[6], Hamideh Ahmadi[7], Elahe Delshad[1]*

[1]Department of Persian Medicine, School of Persian and Complementary Medicine, Mashhad University of Medical Sciences, Mashhad, Iran
[2]Department of Neurosurgical, School of Medicine, Ghaem Hospital, Mashhad University of Medical Sciences, Mashhad, Iran.
[3]Pharmacological Research Center of Medicinal Plants, Mashhad University of Medical Sciences, Mashhad, Iran
[4]Department of Statistics, Macquarie University, Sydney, Australia
[5]Department of Epidemiology and Biostatistics, School of Health, Mashhad University of Medical Sciences, Mashhad, Iran
[6]Department of Neurology, Student Research Committee, School of Medicine, Mashhad University of Medical Sciences, Mashhad, Iran
[7]Department of Neurology, Student Research Committee, School of Medicine, Mashhad University of Medical Sciences, Mashhad, Iran

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Abstract

Experiencing complications within the first two weeks after stroke leads to a high risk of mortality and length of hospitalization. The present pilot study was intended to investigate the hypothesis that adult patients treated with safflower or not would present with fewer neurological complications following 15 days. In a randomized controlled trial, subjects diagnosed with ischemic cerebrovascular accident (CVA) based on focal neurological findings on brain imaging who met the inclusion criteria of our study were recruited from Ghaem Hospital, Mashhad, Iran, between 2016 and 2017. Thirty-six patients were included in the survey and randomly allocated into treatment (A) and control (B) groups. An oral syrup of safflower extract and nasal drop of safflower oil were additionally prescribed for group A. Group B only received a standard anti-ischemic regimen. The primary outcome measure was the National Institutes of Health Stroke Scale score (NIHSS) over 15 days. Safflower treatment led to a notably higher mean difference in the NIHSS score between the baseline score and 15-day post-treatment score in group A in comparison to group B ($p < 0.001$). However, adjustment for covariates (age, gender, and baseline measures) showed no significant reduction in neurological status between them ($p = 0.340$). There was a statistically significant difference in neurological symptom scores between the groups ($p = 0.044$). Based on this pilot study, adjuvant treatment with safflower in addition to the standard anti-ischemic regimen can be more effective than individual conventional drugs for treating ischemic CVA among adults.

Keywords: Traditional medicine; *Carthamus tinctorius*; Stroke; Controlled randomized trial

Introduction

Cerebrovascular diseases are reported as the second cause of death and the first cause of disability across the globe [1]. Stroke is known as the third cause of death following myocardial infarction and cancer worldwide [2]. According to current statistics, there

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*Corresponding Author: Elahe Delshad
Department of Persian Medicine, School of Persian and Complementary Medicine, Mashhad University of Medical Sciences, Mashhad, Iran
Tel: 985138829279
Email: delshade2018@gmail.com

is one death every four minutes due to stroke in the United States of America [3]. Recently, the number of people afflicted with stroke has been on a sharp rise in developing countries [4]. The recent studies have highlighted the growing incidence and mortality rate of stroke among young adults, which, in turn, necessitates the significance of planning and management for the prevention and treatment of this disease [5]. Brain ischemia is one of the common causes of disability in adulthood and is estimated to affect 23 million patients all over the world by 2030. It also induces enormous costs for an individual, as well as the country's health system besides changes in the quality of life and is associated with different adverse effects, including motor paralysis, mental disorders, and even death. Despite the state-of-the-art advances in basic knowledge and therapeutic methods of stroke, its incidence is still increasing [6-8].

These reasons signify the importance and necessity of more efforts for effective treatment in this regard. This cerebrovascular disease develops following insufficient blood flow in the brain. Disturbances in the blood flow may lead to reduced brain function, death of some cells, and stroke [9]. This decline in the cerebral blood flow can arise from thrombosis, arterial emboli, or hemorrhage [9]. From a pathologic view in modern medicine, there are two main mechanisms for stroke: ischemia and hemorrhage. The former mechanism includes emboli, thrombosis, and a decrease in the systemic blood flow [10,11]. Of stroke cases, 85% have been related to ischemic causes [12].

Vascular obstruction causes the interrupting cerebral blood flow in ischemic stroke. Consequently, a complex multistage pathophysiologic process occurs at the cellular and tissue levels, which is known as an ischemic cascade [10,11]. Moreover, a complex of different reactions occurs in neurons including inflammatory pathways, ionic imbalance, apoptosis, and so forth. These mechanisms at two stages induce cellular and tissue impairments. The first stage is ischemic due to reduced blood flow at the target site. The second stage, known as reperfusion, commences due to the return of blood flow from the tissue [13,14]. Ischemic stroke and the subsequent reduction or blockage of local blood flow impose damage and death upon those cells at the core area where blood flow is blocked. On the contrary, those cells at the penumbra also undergo damage and fail to a normal function but are still alive. Therefore, they can be recovered through antioxidant drugs following tissue reperfusion [15].

Accordingly, what is of utmost importance for the effective treatment of disability caused by ischemic stroke is early reperfusion to reduce infarction size after local blood flow blockage [16]. Currently, the most effective therapeutic method is intravenous tissue plasminogen activator (TPA) injection within three hours of symptom onset, which is not possible for all patients [17, 18]. Furthermore, fibrinolytic agents, such as streptokinase and urokinase, are not so effective and have some limitations, adverse effects, and mortality risk [19-21].

Considering the complicated pathophysiology and several mechanisms behind stroke or a cerebrovascular accident (CVA), applying only one therapeutic strategy would not be so effective and thus combining thrombolytic therapy with protective treatments as other potential supplemental alternatives seems more effective [2]. Given the increasing incidence of stroke [23], adverse effects [24], and high cost of physiotherapy and rehabilitation interventions [25], employing supplemental treatments in association with modern medicine is recommended [26]. Alternative therapy based on herbal medicine gives rise to promising results [27]. Herbal agents with relative safety can be used in addition to chemicals with adverse effects and incomplete effectiveness [28]. They not only display synergistic effects with chemicals, but also negate their toxicity consequences [28-30].

In Persian medicine, a subcategory of eastern traditional medicine, some herbs are available that can be associated with modern medicine to effectively treat stroke and its subsequent disability [31]. In the past, physicians knew about cerebrovascular disorders, namely stroke. Most present knowledge and theories are founded on such traditional knowledge [9]. Considering the pathophysiology related to stroke and the mechanisms responsible for clot formation and blockage of blood flow, it is more likely that diluents and anticoagulants mentioned in Persian medicine resources can be utilized as an effective treatment [9]. In the ischemic process, the generation of free radicals increases that culminates in elevated intermolecular oxidation, cellular impairment, and cell death. The use of some herbaceous species and natural products with antioxidant properties can afford to prevent oxidative stress caused by free radicals [32-34]. Of herbs addressed in traditional medicine resources with antioxidant as well as anticoagulant activities, safflower (*Carthamus tinctorius* L.) [35,36], which has three vernacular names in the Persian language, i.e. Golrang, Kajireh, or Kazireh, belongs to the Compositae family [37]. It has been indicated that the dried floret of C. tinctorius known as Carthami flos could treat coronary heart disease, angina pectoris, gynecologic diseases, stroke, and hypertension [38]. Moreover, the effect of red safflower on patients with acute ischemic stroke was studied previously. It was found that red safflower could improve neurologic functional deficits [39]. There are two reports from China highlighting the notable effectiveness of safflower yellow and hydroxysafflor yellow A for acute ischemic stroke against ginkgo leaf and dipyridamole [40,41]. The

lack of evidence from a randomized controlled trial provides the basis for the present study to compare the combined treatment of common anti-ischemic drugs and safflower in patients with ischemic CVA compared to the individual anti-ischemic drugs.

Materials and Methods

Participants

The ethics committee of Mashhad University of Medical Sciences approved this study (IR.MUMS.REC.1394.555). The written consent was taken from all participants after an explanation of goals and interventions for the session. The consolidated standards of reporting trials (CONSORT) guidelines were also adopted to describe the outcomes of this study. The sampling method was convenient. In a single-blind clinical pilot trial among 80 patients referred by the Department of Neurology, Ghaem Hospital in Mashhad, Iran, 36 subjects met the inclusion criteria and were enrolled in the study. Subjects then were randomly divided into the intervention group with 17 patients (group A), and the others (19 cases) were considered as the controls (group B). The sample size was calculated based on the rule of thumb proposed by Browne [42] for the primary outcome (National Institutes of Health Stroke Scale, NIHSS) score between 1 and 24, considering a significance level of 0.05 ($\alpha = 0.05$), a test power of 64%, and drop-outs of 55%.

Patients diagnosed with CVA according to clinical examinations and CT scans were recruited. Indeed, within one week after hospitalization for stroke, they were recruited and then randomized in the two groups. Afterwards, the pertinent questionnaire was filled out for each patient.

Inclusion criteria were focal neurological findings on brain imaging consistent with the diagnosis of ischemic CVA. The following factors would rule out the patients' participation:

Inability to read and sign a written informed consent form, lack of consciousness, intravenous recombinant TPA treatment due to focal neurological symptoms, transient ischemic attack with normal imaging that improves symptoms less than 24 h, NIHSS score with a bad prognosis (> 24), pregnancy (due to the teratogenic side effects of safflower), any evidence of intracranial hemorrhage, taking anticoagulants (i.e., heparin or warfarin), venous infarction following venous sinus thrombosis, chronic systemic diseases, such as chronic liver disease, uremia, and progressive stages of blood disease that might influence our therapeutic protocol or the intervention could aggravate these medical conditions.

The eligible patients were then allocated into groups A and B using a random digit table. Randomization was concealed. The treatment assignment was not blinded. The latter group (B) served as the control group undergoing interventions of common anti-ischemic drugs (modern medicine); while their peers in group A were treated with interventions of traditional/modern medicine. Of 36 patients in this study, 17 were assigned to group A, and the other remaining was considered the control. As many as 7 cases in group A failed to complete the study (Figure 1).

Interventions

Group B received the common anti-ischemic drugs, including aspirin (325 mg/day; Hakim Co., Iran), atorvastatin (20 mg/day; Hakim Co., Iran), and piracetam (800 mg/day; Hakim Co., Iran) in case of low creatinine level for 15 days. Besides, traditional medicine-based therapy in the form of oral syrup of safflower extract and nasal drop of safflower oil was additionally prescribed for group A.

Essential recommendations, treatment instruction, and applicable dosage were explained to the patients both verbally and in writing. Safflower was purchased from Isfahan, Iran and then authenticated by botanist from Mashhad University of Medical Sciences, Mashhad, Iran (Herbarium Number: 4793). The hydroethanolic extract of safflower was prepared and administered in 25% syrup. The patients in group A orally received 5 mL of this syrup every 12 h (that is, 2.5 g/day) over 15 days. A total of 12 g of safflower oil was given in a dose of 3 drops (i.e., 2 g) every 4 h for 15 days. Noteworthy, the administration of safflower below 3 g/day was permitted [43].

A researcher was in charge of patient visits with further control for adherence to the instructions. Fifteen days later, they were directly examined. Telephone calls and clinical examinations were performed considering the participant's age category, using the NIHSS questionnaire at irregular intervals until the end of the treatment. They were allowed to come back for their complications. There was no follow up of the patients.

NIHSS questionnaire quantitatively measures the neurological disorders of stroke, assessing 11 variables, namely level of consciousness, eye movements, visual fields, facial palsy, motor arm, motor leg, ataxia, dysarthria, sensory loss, speech, and inattention [44]. The measurement was performed before and following the intervention. Researchers responsible for data collection and analysis were blind to group allocation and relevant interventions.

Statistical analysis

Given that the sample size was small and attrition might occur because of patients withdrawing from the study, there was a high risk of bias. Data analyses were carried out based on an intention-to-treat to overcome high and unbalanced drop-out rates. Comparison tests, including independent-group t-test and

paired t-test were carried out using Statistical Package for the Social Sciences (SPSS) software. Age, gender, and baseline measures of NIHSS were considered as covariates. A p value of less than 0.05 was considered statistically significant.

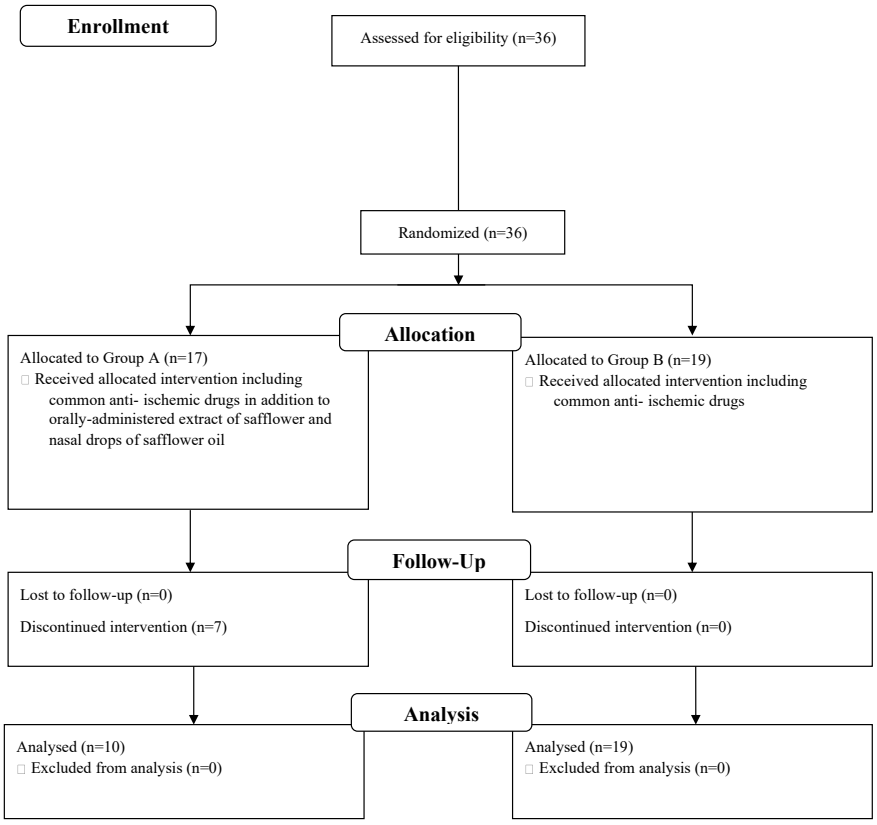

Figure 1. CONSORT flow diagram of the progress through the phases of a one-blind clinical pilot trial of two groups

Results

Of 36 patients recruited in this study, almost half was allocated in group A to receive the traditional medicinal intervention along with conventional treatments. Their counterparts in group B as the controls (52.8%) were instructed to use the modern medicinal treatment. It was demonstrated that the two groups were comparable in terms of age, gender, and clinical characteristics, such as high blood pressure and diabetes ($p > 0.05$) (Table 1). The study also showed a high drop-out rate (19.4%) among these patients. Therefore, the baseline measures of NIHSS, age, and gender had no significant effects on the patients' response to the treatment.

Table 1. Demographic characteristics of the participants at enrolment

Variable	Group A (n = 17)	Group B (n = 19)	p value
Age (Mean ± SD)	72.3 ± 12.0	76.3 ± 9.8	0.10
Males, n (%)	13 (76.5)	12 (63.1)	0.35
High blood pressure, n (%)	10 (58.8)	12 (63.1)	0.31
Diabetes, n (%)	5 (29.4)	4 (21.0)	0.09
Dyslipidemia, n (%)	7 (41.2)	8 (42.1)	0.91
Smoking, n (%)	3 (17.6)	2 (10.5)	0.58
BMI > 30	2 (11.8)	3 (15.8)	0.42

The leading outcome in this study was the NIHSS total score, which evaluates the level of consciousness, eye movements, visual fields, facial palsy, motor arm, motor leg, ataxia, dysarthria, sensory loss, speech, and inattention. The mean scores of the NIHSS at baseline and after 15 days were 12.5 ± 4.6 for the treatment group (A) and 15.0 ± 4.2 for the control group (B) (Table 2). All these subjects were at the moderate severity of NIHSS scores before and following the intervention. It was evident that the higher the score, the more severe the participants' condition. No significant difference was found between the two groups ($p = 0.096$). After the intervention, there was a significant difference between post-scores of the study groups ($p = 0.044$). The mean difference in the NIHSS score between the baseline score and 15-day post-treatment score was considerably higher ($p < 0.001$) in the intervention group (2.0 ± 1.4) than the controls (1.4 ± 1.0). Applying the traditional medicinal intervention was associated with a significant reduction in the patients' neurological status.

Table 2. Comparison of the NIHSS mean scores prior to and following the intervention (Mean ± SD) in this pilot study

Variable	Group A (n = 10)	Group B (n = 19)	p value
Baseline score	12.5 ± 4.6	15.0 ± 4.2	0.096
15-day score	10.5 ± 4.9	13.6 ± 4.1	0.044
Difference	2.0 ± 1.4	1.4 ± 1.0	< 0.001

During the intervention, seven cases of group A failed to complete the study. No adverse effects related to the study treatments were observed in both groups. Of 7 drop-outs in group A, there was only one case of hematuria. Also, two patients died on days 8 and 10. The other four rejected further involvement in the study on days 5, 6, 7, and 10 due to the long distance between the residence place and the Department of Neurology.

Discussion

This pilot study was aimed to investigate the effect of individual conventional medicine or its combination with safflower-based practices on the NIHSS score in patients with ischemic stroke. Our findings indicated combinational therapy significantly decrease the scores of NIHSS than the standard treatment. However, further adjustment for covariates highlighted that the treatment containing safflower was no more effective than that composed of only modern medicine.

Ischemic stroke is a pathologic process involving multiple mechanisms; therefore, an effective treatment should be contained supplemental medicine and herbal agents in addition to the available options. Safflower presented anticoagulant and antioxidant properties with traditional uses for this purpose. Its flower and fruit have potent laxative actions [45]. A total of 104 constituents have been identified in this plant, such as alkaloids, flavonoids, aromatic compounds, and so forth, that cause different biological effects, including dilating coronary artery, improving the immune system, anti-thrombosis, and anticoagulation [45]. Furthermore, safflower can act as an anti-adhesion for platelets, antibacterial agent, anti-inflammation, neuronal protection, and analgesic. Safflower oil also seems to be beneficial for the prevention and treatment of atherosclerosis, besides sedation of rheumatic diseases [46,47]. What is more, its effectiveness for ischemia therapy was a recurrent theme in Persian medicine.

Zakaria Razi described the safflower as a diluent and emollient in his book "*Alhavi*" [48]. In traditional resources, it was stated that "its nature is warm and dry and its strength lasts for three years". As for its characteristics, it was noted that "safflower absolutely melts frozen blood". Also, Hakim Aghili-Khorasani in his book "*Makhzan- Al'Advieh*" maintained that safflower is more effective for medical conditions related to the human head and "a spice melting each frozen phlegm" [49].

Herbal medicine so far has explored various plants for the treatment of ischemia. Safflower, as a candidate for ischemia treatment based on Persian medicine in this study, has not shown promising outcomes for such conditions and proved to be no more effective at therapeutic dosages than the individual treatment using modern medicine. A recent *in vitro* study has revealed that safflower can function as a catalytic agent [50]. HSYA (hydroxyl safflower yellow A), an active agent in safflower, has been reported to be neuroprotective against ischemia, antioxidant, and inhibitory against thrombus formation [51].

Zhu et al., according to the antithrombotic effect of safflower in previous studies, substantiated its neuroprotective functions [52]. In a rat model, Luo et al. demonstrated that safflower could afford to cause a dose-dependent reduction of infarction size in ischemic stroke; put differently, the high concentration of safflower led to a more decreased size [53].

Administering safflower extract after ischemic stroke in mice, Zhao et al. indicated its lowering impact on ischemia-induced damage and also its protection on the brain [54]. Likewise, Yu-Ye et al. observed that safflower considerably diminished cerebral edema caused by global cerebral ischemia in the case group compared with the controls [55]. Wei et al. developed a model of ischemia and middle artery occlusion in rats and measured chemical neurotransmitters in response to safflower. The study demonstrated the antioxidant and neuroprotective effects of safflower against ischemia, and its capability to attenuate infarction size [56]. Another similar study by Ye et al. showed that HSYA mitigated the inflammatory reactions following ischemia [51]. A model of blood stasis in rats was developed by Li et al. to examine the effectiveness of carthamins yellow (CY) extracted from safflower. It was shown that CY exerted a decreasing impact on the whole blood and plasma viscosity, erythrocyte aggregation index, hematocrit, and platelet aggregation. On the other hand, CY elevated blood fluidity and induced anticoagulation to some extent [57].

Treating rats exposed to middle cerebral artery occlusion and reperfusion with HSYA unraveled amelioration of blood-brain barrier disruption and brain edema [58]. Safflower was observed to protect neurons against ischemia–reperfusion injury through antioxidant activity and inhibition of thrombin production [59]. An animal study by Wu et al. indicated that safflower extract at the concentrations of 20, 40, and 80 mg/kg and aspirin 5 mg/kg increased thrombosis occlusion time. As for the weight of thrombus, safflower was more effective than aspirin [60]. Moreover, safflower extract can reduce platelet aggregation caused by adenosine diphosphate and blood coagulation. Despite the disparity between toxic and therapeutic doses, its application should be avoided during pregnancy and lactation [61]. Such beneficial effects of safflower on stroke have been also addressed in systematic studies [62]. More recently, a rat model of ischemia–reperfusion brain injury exposed to safflower presented a decrease in the infarction area and neurological deficits. It can afford to lower blood levels of free radicals, as well as the expression of tumor necrosis factor-α and interleukin-1β [63].

All positive outcomes have been found through *in vitro* and in *vivo* studies. Therefore, further efforts on a human model are required to elucidate the effectiveness of safflower on stroke. The therapeutic dosage of safflower extract is 3 g daily with no noticeable side effects or limitations [64]. Other resources reported 20-40 g of safflower seed as an allowable dose [45].

This study was initially conducted on 36 patients, of whom 19 persons were allocated in the control group (B). As for the intervention group (A), seven failed to complete the intervention. Due to the small sample size, the generalization of these findings needs a larger one and more prolonged follow-up. Another limitation of the present study regards the participant's initial severity of stroke which mostly included moderate symptoms. Therefore, it is less likely to apply this therapeutic dose for severe symptoms (NIHSS score > 24).

Following the acute phase, patients afflicted with stroke improve to some extent in limb movement, verbal response, and so forth either spontaneously or via rehabilitation interventions, such as physiotherapy, occupational therapy, and speech therapy that are regarded as a part of the stroke natural course, and are attributed to regeneration and proliferation of the cells at the penumbra. Therefore, NIHSS scores are recommended to be measured during long follow-ups and compared between the two groups to determine the specific effects of this treatment. The limitation of research on safflower is related to its formulation. Safflower in the form of the oral syrup has considerable coloring effects, which were unpleasant for patients; therefore, it should be provided in a capsule form. The standard treatment for stroke within 4.5 h of symptom onset is antiplatelet drugs, including aspirin. It would be in the interest of the reader to further understand whether safflower intervenes with such conventional therapies and affects their effectiveness biochemically.

Conclusion

The present study indicated that the combination of safflower-based practices with modern therapy could be more effective than individual modern medicine for reducing the NIHSS scores of ischemic stroke patients. Further trials using a large sample size using different type of safflower extract and dosage are suggested for future studies.

Conflict of Interests

None.

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References

[1] Marini C, Russo T, Felzani G. Incidence of stroke in young adults: a review. Stroke Res Treat 2011;2011: 535672.

[2] Lloyd-Jones D, Adams RJ, Brown TM, Carnethon M, Dai S, et al. Heart disease and stroke statistics--2010 update: a report from the American Heart Association. Circulation 2010;121:e46-e215.

[3] Benjamin EJ, Blaha MJ, Chiuve SE, Cushman M, Das SR,

et al. Heart disease and stroke statistics—2017 update: a report from the american heart association. Circulation 2017;135:e146-e603.

[4] Venketasubramanian N, Yoon BW, Pandian J, Navarro JC. Stroke epidemiology in south, east, and south-east asia: a review. J Stroke 2017;19:286-294.

[5] Krishnamurthi RV, Moran AE, Feigin VL, Barker-Collo S, Norrving B, et al. Stroke prevalence, mortality and disability-adjusted life years in adults aged 20-64 years in 1990-2013: data from the global burden of disease 2013 study. Neuroepidemiology 2015;45:190-202.

[6] Guan T, Ma J, Li M, Xue T, Lan Z, et al. Rapid transitions in the epidemiology of stroke and its risk factors in China from 2002 to 2013. Neurology 2017:89:53-61.

[7] Swanepoel AC, Pretorius E. Prevention and follow-up in thromboembolic ischemic stroke: do we need to think out of the box? Thromb Res 2015;136:1067-1073.

[8] Tavakoli Rezaee H, Kamrani N. The effect of ascorbic acid on infarction volume and neurological disorders following stroke of embolic model in rats. J Rafsanjan Univ Med Sci 2009;8:49-58.

[9] Zargaran A, Zarshenas MM, Karimi A, Yarmohammadi H, Borhani-Haghighi A. Management of stroke as described by Ibn Sina (Avicenna) in the Canon of Medicine. Int J Cardiol Heart Vasc 2013;169:233-237.

[10] Omidi R, Zali H, Rezaee TM, Modara F. Investigation of ischemic stroke mechanism by analyzing human brain proteome. J Ilam Uni Med Sci 2013;21:98-108.

[11] Attari F, Karimzadeh F, Gorji A. Acute ischemic stroke: Pathophysiology and stem cell therapy. J Paramed 2013;7:46-51.

[12] Giele JL, Witkamp TD, Mali WP, van der Graaf Y. Silent brain infarcts in patients with manifest vascular disease. Stroke 2004;35:742-746.

[13] Xing C, Arai K, Lo EH, Hommel M. Pathophysiologic cascades in ischemic stroke. Int J Stroke 2012;7:378-385.

[14] Pan J, Konstas A-A, Bateman B, Ortolano GA, Pile-Spellman J. Reperfusion injury following cerebral ischemia: pathophysiology, MR imaging, and potential therapies. Neuroradiology 2007; 49:93-102.

[15] Rahnema M, Foroozandeh M, Ghasemloo E. The effect of hydroalcoholic extract of Satureja hortensis on outcomes of stroke in rats. Feyz 2016;19:511-519.

[16] Juttler E, Kohrmann M, Schellinger PD. Therapy for early reperfusion after stroke. Nat Clin Pract Cardiovasc Med 2006;3:656-663.

[17] Cheng NT, Kim AS. Intravenous Thrombolysis for acute ischemic stroke within 3 hours versus between 3 and 4.5 hours of symptom onset. Neurohospitalist 2015;5:101-109.

[18] Fu Y, Wang Y, Zhang B. Systems pharmacology for traditional Chinese medicine with application to cardio-cerebrovascular diseases. J Tradit Chin Med Sci 2014;1:84-91.

[19] Behravan E, Razavi BM, Hosseinzadeh H. Review of plants and their constituents in the therapy of cerebral ischemia. Phytother Res 2014;28:1265-1274.

[20] Chen YF. Traditional Chinese herbal medicine and cerebral ischemia. Front Biosci 2012;4:809-817.

[21] Naderi G, Jaefari Dinani N, Nejabat N, Kelardasht M, Jaefarian Dehkordi A, et al. Fibrinolytic activity of the polyphenolic extract of Curcuma domestica Valet., Cinnamomum verum J. Presl, Heracleum persicum Desf. Ex Fischer, and Zataria multiflora Boiss. in in vitro. Iranian Journal of Medicinal and Aromatic Plants Research 2011;4:572-580.

[22] Derex L, Paris C, Nighoghossian N. Combining intravenous thrombolysis and antithrombotic agents in stroke: an update. J Am Heart Assoc 2018;7:54-74.

[23] Wafa HA, Wolfe CDA, Rudd A, Wang Y. Long-term trends in incidence and risk factors for ischaemic stroke subtypes: prospective population study of the south london stroke register. PLoS Med 2018;15:26-49.

[24] Bansal S, Sangha KS, Khatri P. Drug treatment of acute ischemic stroke. Am J Cardiovasc Drugs 2013;13:57-59.

[25] Tummers JFMM, Schrijvers AJP, Visser-Meily JMA. Economic evidence on integrated care for stroke patients; a systematic review. Int J Integr Care 2012;12:1-15.

[26] Pan W, Yang X, Zhou H. The development of integrative medicine. Integr Med Int 2016;3: 96-98.

[27] Gaire BP. Herbal medicine in ischemic stroke: challenges and prospective. Chin J Integr Med 2018;24:243-246.

[28] Zhang J, Onakpoya IJ, Posadzki P, Eddouks M. The safety of herbal medicine: from prejudice to evidence. Evid -Based Complement Altern Med 2015;2015:316706.

[29] Chan TY. Interaction between warfarin and danshen (Salvia miltiorrhiza). Ann Pharmacother 2001;35:501-504.

[30] Izzo AA. Interactions between herbs and conventional drugs: overview of the clinical data. Med Princ Pract 2012;21:404-428.

[31] Kim H. Neuroprotective herbs for stroke therapy in traditional eastern medicine. Neurol Res 2005;27:287-301.

[32] Sinha K, Chaudhary G, Gupta YK. Protective effect of resveratrol against oxidative stress in middle cerebral artery occlusion model of stroke in rats. Life Sci 2002;71:655-665.

[33] Wang Q, Simonyi A, Li W, Sinnenesk BA, Miller RL, et al. Dietary grape supplement ameliorates cerebral ischemia-induced neuronal death in gerbils. Mol Nutr Food Res 2005;49:443-451.

[34] Jiang J, Wang W, Sun YJ, Hu M, Li F, et al. Neuroprotective effect of curcumin on focal cerebral ischemic rats by preventing blood-brain barrier damage. Eur J Pharmacol 2007;561:54-62.

[35] Yao D, Wang Z, Miao L, Wang L. Effects of extracts and isolated compounds from safflower on some index of promoting blood circulation and regulating menstruation. J Ethnopharmacol 2016;191:264-272.

[36] Zhou X, Tang L, Xu Y, Zhou G, Wang Z. Towards a better understanding of medicinal uses of Carthamus tinctorius L. in traditional Chinese medicine: a phytochemical and pharmacological review. J Ethnopharmacol 2014;151:27-43.

[37] Delshad E, Yousefi M, Sasannezhad P, Rakhshandeh H, Ayati Z. Medical uses of carthamus tinctorius L. (Safflower): a comprehensive review from traditional medicine to modern medicine. Electron Physician 2018;10:6672-6681.

[38] Tu Y, Xue Y, Guo D, Sun L, Guo M. Carthami flos: a review

of its ethnopharmacology, pharmacology and clinical applications. Rev Bras Farmacogn 2015;25:553-566.
[39] Ma L, Li D, Li K. Clinical effect study on ischemic stroke in acute period treated with red flower injection. J Zhejiang Chin Med Univ 2012;36:633-635.
[40] Xiao X, Hu W. Clinical effect of HSYA injection on acute ischemic apoplexy. Med J West China 2011;33:932-936.
[41] Xing Y, Tu Q, Xiao Q, Jiao Y. Clinical effect of safflower yellow injection on ischemic apoplexy. Chin Hosp Pharm J 2008;28:1493-1495.
[42] Browne RH. On the use of a pilot sample for sample size determination. Stat Med 1995;14:1933-1940.
[43] Imami A, Taleb A, Khalili H. PDR for Herbal Medicines. 1st ed. Andisheh-Avar. Tehran 2010.
[44] Brott T, Adams HP, Olinger CP, Marler JR, Barsan WG, et al. Measurements of acute cerebral infarction: a clinical examination scale. Stroke 1989;20:864-870.
[45] Zargari A. Mdical Herbs. Tehran University Press. Tehran 2012.
[46] Fattahi M, Nobakht M, Mahmoudian M. The teratogenic effect of Golrang's extract on development of CNS in mouse. Razi Journal of Medical Sciences 2000;7:138-143.
[47] Mozafarian V. Recognition of Medicinal and Aromatic Herbs in Iran. Farhang-e-Moaser. Tehran 2012.
[48] Zakaria al-Razi M. Al-Havi fi al-Teb. Dar al-Hayat al-Torath al-Arabi. Beirut 2001.
[49] Aghili Khorasani MH. Makhzan Aladviyeh. Sabz-arang Press. Tehran 2012.
[50] Yousefi M, Rakhshandeh H, Delshad E, Mahjoub F. Assessment of clot lytic effect of Carthamus tinctorius (Golrang). Avicenna J Phytomed 2015;5:78. 1/2p.
[51] Ye S-Y, Gao W-Y. Hydroxysafflor yellow A protects neuron against hypoxia injury and suppresses inflammatory responses following focal ischemia reperfusion in rats. Arch Pharm Res 2008;31:1010-1015.
[52] Zhu H, Wang Z, Ma C, Tian J, Fu F, et al. Neuroprotective effects of hydroxysafflor yellow A: in vivo and in vitro studies. Planta Med 2003;69:429-433.
[53] Luo J, Fang Z, Zhou L, Lai S. Effects of carthamus tinctorius injection on bcl-2, caspase-3 expression related to neurons apoptosis after local cerebral ischemia. Zhongguo Zhong Yao Za Zhi 2004;29:977-980.
[54] Zhao Q, Du J-S, Han X-m, Xu F, Yang X, et al. Research of apotosis after acute cerebral ischemia reperfusion in rat brain and safflower protection mechanism. Chin J Lab Med 2004;8:006.
[55] Yu-Ye X, Yang M, Sheng Y-C. Neuroprotective effect of hydroxysafflor yellow A on cerebral ischemia injuries in rats. Chinese Journal of Pharmaceuticals. 2005;36:1-18.
[56] Wei X, Liu H, Sun X, Fu F, Zhang X, et al. Hydroxysafflor yellow A protects rat brains against ischemia-reperfusion injury by antioxidant action. Neurosci Lett 2005;386:58-62.
[57] Li H-X, Han S-Y, Wang X-W, Ma X, Zhang K, et al. Effect of the carthamins yellow from Carthamus tinctorius L. on hemorheological disorders of blood stasis in rats. Food Chem Toxicol 2009;47:1797-802.
[58] Sun L, Yang L, Xu Y-W, Liang H, Han J, et al. Neuroprotection of hydroxysafflor yellow A in the transient focal ischemia: inhibition of protein oxidation/nitration, 12/15-lipoxygenase and blood–brain barrier disruption. Brain Res 2012;1473:227-235.
[59] Chen L, Xiang Y, Kong L, Zhang X, Sun B, et al. Hydroxysafflor yellow A protects against cerebral ischemia–reperfusion injury by anti-apoptotic effect through PI3K/Akt/GSK3β pathway in rat. Neurochem Res 2013;38:2268-2275.
[60] Wu S-H, Zheng C-P, Chen S-Y, Cai X-P, Shi Y-J, et al. Anti-thrombotic effect of carthamus tinctorius linn extracts in rats. Trop J Pharm Res 2014;13:1637-1642.
[61] Namjoo A, Rafieian M, Azizi S, Talebi-Juneghani A. Histopathologic effects of carthamus tinctorius on the brain liver and kidney of the new born mice. J Shahrekord Univ Med Sci 2010; 11:38-45.
[62] Fan S, Lin N, Shan G, Zuo P, Cui L. Safflower yellow for acute ischemic stroke: a systematic review of randomized controlled trials. Complement Ther Med 2014;22:354-361.
[63] Fu P-K, Pan T-L, Yang C-Y, Jeng K-C, Tang N-Y, et al. Carthamus tinctorius L. ameliorates brain injury followed by cerebral ischemia-reperfusion in rats by antioxidative and anti-inflammatory mechanisms. Iranian jouenrnal of basic medical sciences. 2016;19:1368-1375.
[64] Heber D. PDR for herbal medicines. Montvale. Thomson 2004.